SYNTHETIC STUDIES ON THE C-RING OF GIBBERELLINS IN RELATIONSHIP TO THE ANTHERIDIOGENS FROM THE FERNS OF THE FAMILY SCHIZAEACEAE

A Thesis Submitted for

the Degree of

Doctor of Philosophy

of

The Australian National University

Research School of Chemistry

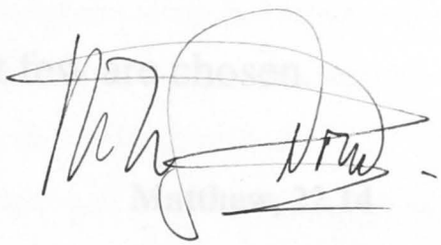
by

Milan Pour, M.Sc.

September 1994

DECLARATION

This Thesis contains no material previously submitted for a degree in any other University and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference is made in the text. As far as possible, established methodology has been acknowledged by citations of the original publications.



Milan Pour

Many are called, but few are chosen.

Matthew, 22.14

ACKNOWLEDGEMENTS

I am deeply indebted to Professor Lew Mander for guiding me through the Kingdom of synthetic organic chemistry and for his support. The opportunity to work with him and his friendship have meant a great deal to me.

The help of the ANU NMR centre staff over my three years at the University has been very much appreciated. In particular to Max Keniry, Peta Simmons, Chris Blake, Tin Culnane and Barry Gray, who were always willing to teach me how to record and process the results of more sophisticated two-dimensional experiments. I should also like to thank Reet Bergmann from the Analytical Unit and Jenny Rothschild from the Mass Spectrometry Unit for running my samples immediately when I was in urgent need of the results. X-ray structural analyses were performed by Dr. Tony Willis.

I am grateful for all the effort the secretarial staff at the Research School of Chemistry have given me at times when I needed to settle any matters associated with the alien world outside the laboratory. The administrative skills of Penny Richardson, Maureen Slocum, Dawn Walters and Leonie Hoorweg were of great help, especially in dealing with the central administration at the ANU.

I doubt that my time in Canberra could have been enjoyable or rewarding without the support of few close friends. I am very thankful to Jonathan Morris, David Camp, Stephan Kohlstruk, Gerald Haaima, Bruce Twitchin and Barbara Frey for numerous discussions and their generous friendship. I am also grateful to Michael Sherburn for helpful discussions about chemistry.

A big thank-you goes to Alex and Zlata Wallner, Julius and Jana Andres and Kylie Catchpole for making Canberra feel more like home. Julius in particular must be acknowledged for keeping me in touch with Czech humour and Czech-style life attitudes.

My special thanks must go to Stephan Kohlstruk, John Lambert and David Camp for their proof-reading efforts and willingness to be involved in the process of reducing the number of mistakes in this thesis to minimum.

I should like to express my gratitude to the Commonwealth of Australia for the award of an Overseas Postgraduate Research Scholarship and to the Australian National University for the award of an ANU PhD Scholarship.

The gratitude I feel towards my family is difficult to express. I want to thank my parents in particular, as they have given so much and asked so little in return.

ABSTRACT

This thesis describes the partial synthesis of 9,15-cyclogibberellins hydroxylated in the C-ring. The first chapter describes previous biological studies on the biosynthesis of antheridic acid from a 9,15-cyclogibberellin precursor. As a consequence of these experiments, some new compounds, tentatively assigned as 9,15-cyclogibberellins bearing a hydroxy group in the C-ring, were isolated. The discovery of hitherto unknown natural antheridiogen and the subsequent GC-MS analysis leading towards its tentative assignment as 3,12-dihydroxy-9,15-cyclo-GA_9 is likewise discussed. The concluding section of this Chapter contains a brief survey of various strategies, which served as a background to the synthetic endeavours attempted during the preparation of these unusual 9,15-cyclo GAs.

The synthesis of the 11-hydroxy series of derivatives is described in Chapter 2. Following the 19,10 → 19,2 A-ring lactone reversion in a commercially available derivative, functionality was extended from the A-ring into the C-ring by means of an allylic bromination. This process introduced a synthetic function at C(11) and set the stage for the closure of the cyclopropyl ring. The synthesis was then completed in a straightforward manner by functional group interconversions.

Chapter 3 describes two approaches towards the 12-hydroxy series of compounds. In the first synthesis, the 16,17-double bond in the D-ring is functionalised to afford a methylene group with a hydroxy substituent. Consequently, a hydroxy group could be introduced at C(12) by the remote site functionalisation reaction with $Ph(IOAc)_2$. Another sequence of reactions was then employed to close the cyclopropyl ring.

The second approach, based on a formal transposition of the hydroxy group from C(11) to C(12), utilizes the ready availability of the 11-hydroxy compounds. The 11-OH group was eliminated and thus obtained 11,16-diene compound treated with borane-dimethyl sulfide complex to afford, after oxidative work-up, a 12β,17-dihydroxy

derivative. As an attempted protection/ deprotection/ elimination sequence aimed at removing the 17-hydroxy group and restoring the 16,17-double bond presented a difficult task due to the participation of the 12β-hydroxy function, the configuration at C(16) was inverted through enolisation of the 17-oxo function prior to the completion of the synthesis. A full set of four diastereomers of 3,12-dihydroxy-9,15-cyclo-GA_9 methyl ester and two epimers of 12-hydroxy-9,15-cyclo-GA_9 methyl ester was prepared *via* this route.

Chapter 4 describes the synthesis of a new antheridiogen recently isolated from the fern, *Lygodium Japonicum*, which is structurally related to 12-hydroxy-9,15-cyclo compounds. This compound, tentatively assigned as 12-hydroxy-GA_{73}, possessed the same hydroxylation pattern and the 9(11)-ene function, isomeric to the 9,15-cyclo arrangement. Following an improved preparation of a previously described 9(11)-ene compound, the desired goal was achieved by an allylic functionalisation process and functional group interconversions.

TABLE OF CONTENTS

ABBREVIATIONS

The following abbreviations have been used throughout this thesis:

Ac	Acetyl
AIBN	Azobisisobutyronitrile
APT	Attached Proton Test
Bu	Butyl
t-Bu	*tert*-Butyl
BSTFA	Bis(trimethylsilyl)trifluoroacetamide
DAIB	(Diacetoxyiodo)benzene
DBU	1,8-Diazabicyclo[5.4.0]undec-7-ene
DEPT	Distortionless Enhancement by Polarisation Transfer
DIPEA	*N,N*-Diisopropylethylamine
DMAP	4-Dimethylaminopyridine
DMF	*N,N*-Dimethylformamide
DQF COSY	Double Quantum Filtered Correlation Spectroscopy
EI	Electron Impact
Et	Ethyl
eq.	equivalent(s)
GA_n	Gibberellin A_n
GAs	Gibberellins
GC-MS	Gas Chromatography - Mass Spectrometry
HETCOR	Heteronuclear Correlation Spectroscopy
LR HETCOR	Long Range Heteronuclear Correlation Spectroscopy
HMQC	Heteronuclear Multiple Quantum Coherence
HPLC	High Performance Liquid Chromatography
HRMS	High Resolution Mass Spectrometry
IR	Infrared
J	Coupling Constant
KRI	Kovats Retention Index

$M^{\cdot+}$	Molecular Ion
Me	Methyl
MEM	2-Methoxyethoxymethyl
MOM	Methoxymethyl
Ms	Mesyl
MTM	Methylthiomethyl
m/z	Mass To Charge Ratio
NBS	*N*-bromosuccinimide
NMR	Nuclear Magnetic Resonance
2D NMR	Two Dimensional Nuclear Magnetic Resonance Spectroscopy
Ph	Phenyl
ppm	Parts per Million
TBDMS	*t*-Butyldimethylsilyl
TFAA	Trifluoroacetic Anhydride
THF	Tetrahydrofuran
TLC	Thin Layer Chromatography
TMS	Trimethylsilyl
TOCSY	Total Correlation Spectroscopy
Tos	*p*-Toluenesulfonyl
R_f	Retardation Factor
SEM	2-(Trimethylsilyl)ethoxymethyl
UV	Ultraviolet

CHAPTER 1

1. INTRODUCTION

1.1 GENERAL INTRODUCTION

1.1.1 Phytohormones in the process of plant evolution

A most important role in the development of living organisms is played by chemical signals, which, in a given species, at a specific time in ontogenesis, and in a specific manner, determine the correct development of form and structure. Accordingly, the history of phylogenesis of form and structure is closely associated with the history of evolution of hormonal molecules. In contrast to animal hormones, which often acquired a signal function immediately after their first appearance in evolution, most phytohormones, such as abscisic acid, cytokinins, ethylene and to some extent even gibberellins, are ancient reaction products of general metabolism. Most of them are already present in prokaryotic organisms and they have secondarily acquired the character of a developmental signal. Unlike animal evolution, the phylogeny of plants seems to be characterized by a post-evolution of the hormone receptor molecule. This event was a prerequisite for a common and ubiquitously distributed metabolic product to take over the function of a regulatory substance[1].

The current amount of knowledge about the phytohormone receptor proteins is considerably limited and for this reason, only the most primitive taxonomical groups in which a given hormone demonstrates for the first time a signal function for differentiation and morphogenesis can be analyzed[1]. The family of *Schizaeaceous* ferns is a distinct example of a group of lower plants which is under investigation with regard to the evolutionary aspects of the biosynthesis and the biological role of phytohormones in the plant Kingdom.

1.1.2 Regulation of the sexual reproduction of ferns

If the spore of a heterosporous fern is given a chance to germinate, it produces a gametophyte plant, which usually takes the form of a small green, heart-shaped structure called the prothallus[2]. The prothallia are completely independent of the large spore-bearing fern plant[3]. Their function is to produce the distinct male and female sexual organs, termed antheridia and archegonia respectively. The formation of antheridia usually precedes archegonium initiation; both organs can be found on the underside of a prothallus[2]. Male sperm liberated from the globular antheridia swim to and pass down the neck of the flask-like archegonium and fuse with the egg at its base. A new spore-bearing plant develops from this fertilized egg.

In 1950 Döpp demonstrated[4,5] that the formation of antheridia on young prothallia of *Pteridium aquilium* (bracken fern) and *Dryopterix filix-mas* (male fern) was induced by a substance which could be extracted from mature prothallia. Following this discovery, several discrete compounds, for which the term antheridiogen has been coined, have been isolated from the gametophytes of other fern species[6]. It was observed for the members of the family *Schizaeaceae* that these substances possessed gibberellin-like activity[7] and, conversely, that gibberellins had antheridium-inducing properties[8]. Nakanishi *et al.* used this information in arriving at formula **1**[9] for antheridic acid[10], the major antheridiogen from *Anemia phyllitidis*. This structure was later revised to **2** (Figure 1) following total syntheses of the respective racemates by Corey and Myers[11].

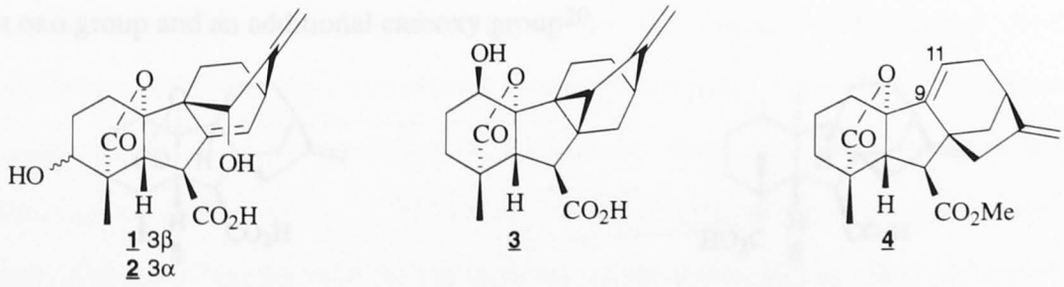


Figure 1. Structural types of natural antheridiogens

Antheridic acid **2** has also been shown to be a natural antheridiogen in other members of the *Anemia* genus, i.e. *A. hirsuta*[12], *A. rotundifolia*, and *A. flexuosa*[13]. It could not be detected in *A. mexicana*, but a new gibberellin-like antheridiogen was obtained from this last species[14], for which structure **3** was deduced[15] and confirmed by synthesis from gibberellin A_7[16]. Structures of two antheridiogens from the related genus *Lygodium japonicum* have also been elucidated, the more potent of which was shown to be 9,11-didehydro-GA_9 methyl ester (**4**)[17,18]. Generally, antheridiogens induce antheridia formation at concentrations as low as 10^{-14} M, and also initiate spore germination, but at higher concentrations (10^{-11} - 10^{-10} M)[29].

1.1.3 Gibberellins in higher plants

Various phases of higher plant development, such as seed germination, the breaking of dormancy, enzyme synthesis, reversal of dwarfism, induction of stem growth, stimulation of flowering, modification of flower sex expression, parthenocarpic development of fruit, fruit enlargement and inhibition of senescence are regulated by a distinct group of plant hormones called the gibberellins[19]. They presently form a group of over 90 highly functionalized diterpenoids which are widely distributed in higher plants. The basic *ent*-norgibberellane formula is constant for ca. 60 metabolites, for which GA_9 (gibberellin A_9, **5**) may be regarded as the parent structure. The differences in constitution are largely accounted for by the location and number of hydroxy groups (up to four). Further variations include an additional double bond, an epoxy function, an oxo group and an additional carboxy group[20].

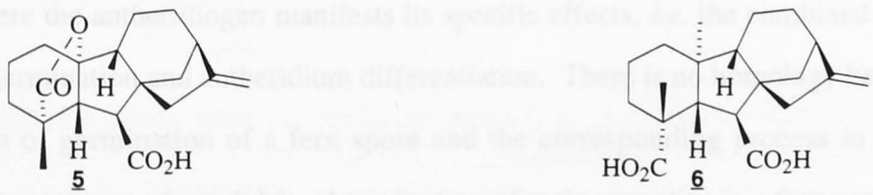


Figure 2. Representatives of the structural types of gibberellins

Most of the remaining GAs possess the full 20-carbon ent-gibberellane skeleton, reflecting their origin from geranylgeranyl pyrophosphate. The parent compound is GA_{12} (**6**) and for the most part, further variations in structure for this group of C-20 gibberellins stem from the level of oxidation at C(20) and/or the addition of one or more hydroxy groups[20].

1.1.4 Antheridiogens in relation to gibberellins

In terms of structure, there is a striking similarity between antheridiogens and gibberellins. A number of observations and assumptions supporting the idea of a close relationship between antheridiogens in *Schizaeaceae* and gibberellins in higher plants have been made over the past twenty years[1].

A representative comparison may be made based on a part of the biological cycle of the *Anemia* genus[1]. The *Anemia* spore population can be divided into two groups. One group, lying on the surface, germinates immediately after receiving optimal humidity conditions and gives rise to the typical two-dimensional gametophyte. The second population, covered by debris, remains ungerminated because of the requirement for light. It has been found that it is this population which is the main target of the antheridiogen signal. Laboratory experiments showed that each single gametophyte which develops on the surface is, as a consequence of pheromone synthesis and secretion, surrounded by a halo of antheridiogen. In the case of cultures on agar, this halo often has a radius of more than 30 cm. Even on natural substrates, a hemisphere of high antheridiogen activity with a 10 cm radius can be formed. In this hemisphere the antheridiogen manifests its specific effects, *i.e.* the combined induction of dark germination and antheridium differentiation. There is no homology between the induction of germination of a fern spore and the corresponding process in a seed as such. The same may be valid for the induction of male sexuality in a fern gametophyte and a flower apex. If it is accepted though, that this receptor/signal system of *Schizaeaceous* ferns was transferred during evolution into spermatophytes, whereby the pheromone function changed to that of a hormone, then several gibberellin reactions of

higher plants can be rationalized in terms of this evolutionary development. This is particularly the case for the phenomenon of dark induction in the germination of several seeds by gibberellins, a reaction which does not happen in natural substrates because the gibberellin concentration is too low. This and a number of similar observations led Schraudolf to the suggestion[1] that the action of antheridiogens in *Schizaeaceous* ferns represents the "moment of becoming a hormone" for gibberellin-like molecules in higher plants.

Investigation into the biosynthesis of both antheridiogens and gibberellins can undoubtedly bring further evidence either for or against this hypothesis and discover the biosynthetic relations between these two classes of compounds. It should be pointed out, however, that the evolutionary link between the *Schizaeaceous* pheromone reaction and the gibberellin responses of higher plants cannot be definitely proved until a direct correlation of their binding proteins has been made.

1.1.5 Biosynthesis of antheridic acid

In the original paper[9] on the structure of antheridic acid (**2**), Nakanishi *et al.* suggested that this compound may well have been formed biogenetically by the rearrangement of a 9,10-epoxide (Figure 3).

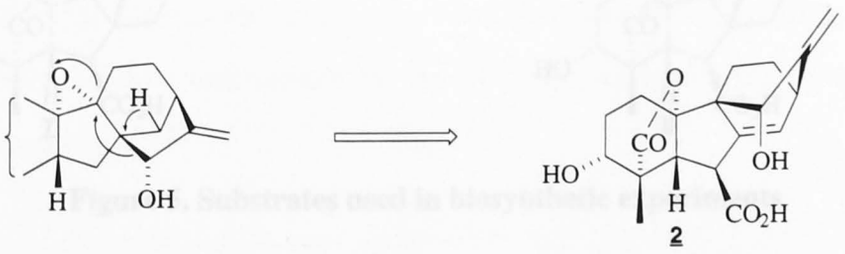


Figure 3. Biosynthesis of antheridic acid 2 as proposed by Nakanishi

The occurrence of three skeletal types, i.e. **2**, **3** and **4** in the family of *Schizaeaceous* ferns, however, led Yamane and Mander to the speculation that there was a close biosynthetic relationship among them. These initial thoughts culminated in the

formulation of a hypothesis[18] that the ent-9,15-cyclogibberellane structure could be a precursor to either antheridane (pathway c) or ent-9,11-didehydrogibberellane derivatives (pathway a), or possibly both. Alternatively, a C(9) cationic intermediate could be a common precursor to each of the three classes of compounds (Figure 4).

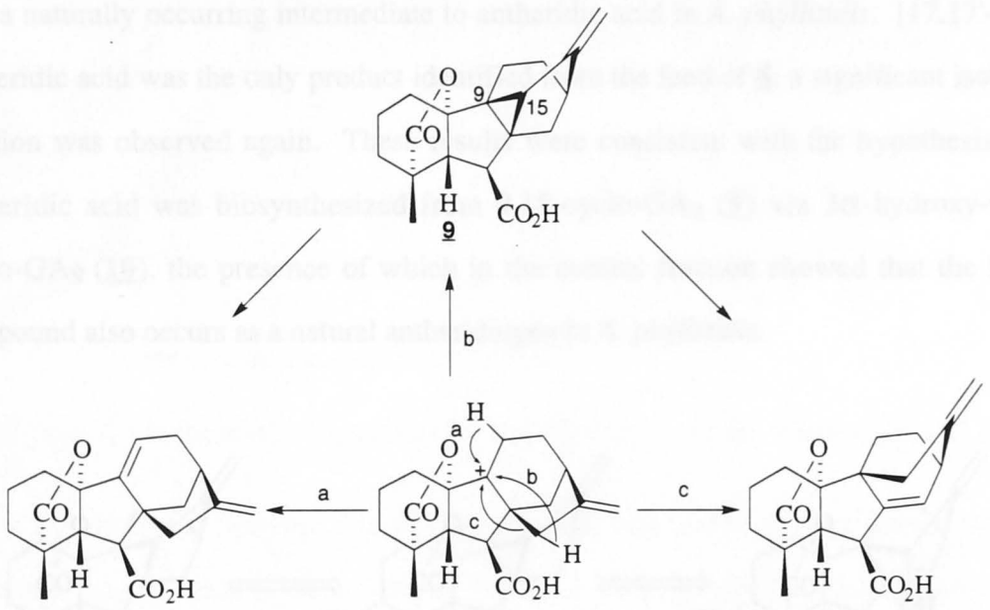


Figure 4. Possible biosynthetic relationship among antheridiogens

Following this proposal, Mander *et al.* synthesized deuterium-labelled compounds **7** and **8**[21](Figure 5).

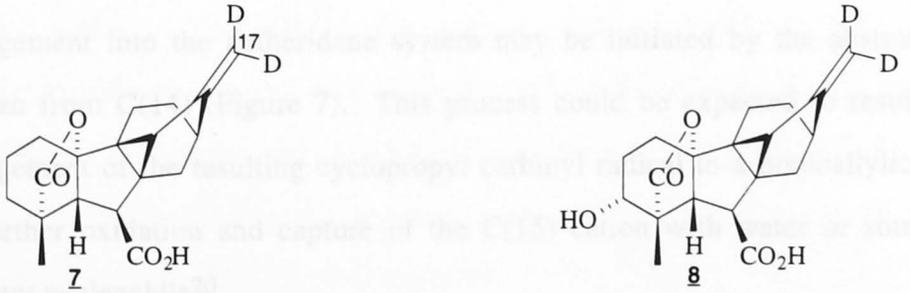


Figure 5. Substrates used in biosynthetic experiments

Both derivatives were fed to prothallia of the fern, *Anemia phyllitidis* and the metabolites from these feeds were subjected to full-scan GC-MS[21] (after treatment with diazomethane to convert the acids into the methyl esters and BSTFA to silylate the hydroxy groups). The outcome clearly indicated that [17,17'-$^2H_2$] 9,15-cyclo-GA_9 (**7**) was converted into [17,17'-$^2H_2$] 3α-hydroxy-9,15-cyclo-GA_9 (**8**), [17,17'-$^2H_2$]

antheridic acid ([17,17'-$^2H_2$]-**2**), [17,17'-$^2H_2$] 1β -hydroxy-9,15-cyclo-GA$_9$ ([17,17'-$^2H_2$]-**3**) and two unknown [$^2H_2$] monohydroxy-9,15-cyclo-GA$_9$-like compounds. Isotopic dilutions of [17,17'-$^2H_2$] 3α-hydroxy-9,15-cyclo-GA$_9$ and [17,17'-$^2H_2$] antheridic acid suggested that 3α-hydroxy-9,15-cyclo-GA$_9$ (**10**) (Figure 6) was a naturally occurring intermediate to antheridic acid in *A. phyllitidis*. [17,17'-$^2H_2$] antheridic acid was the only product identified from the feed of **8**; a significant isotopic dilution was observed again. These results were consistent with the hypothesis that antheridic acid was biosynthesized from 9,15-cyclo-GA$_9$ (**9**) via 3α-hydroxy-9,15-cyclo-GA$_9$ (**10**), the presence of which in the control fraction showed that the latter compound also occurs as a natural antheridiogen in *A. phyllitidis*.

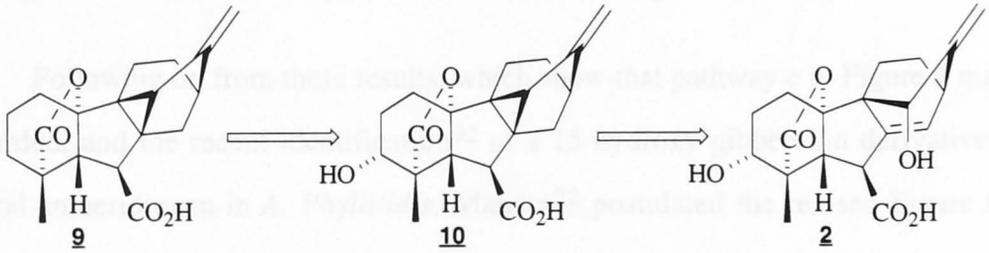


Figure 6. Intermediates in the biosynthesis of antheridic acid

It appears possible that after hydroxylation at C(3) to produce **10**, the rearrangement into the antheridane system may be initiated by the abstraction of hydrogen from C(14) (Figure 7). This process could be expected to result in the rearrangement of the resulting cyclopropyl carbinyl radical to a homoallylic system with further oxidation and capture of the C(15) cation with water or some other equivalent nucleophile[20].

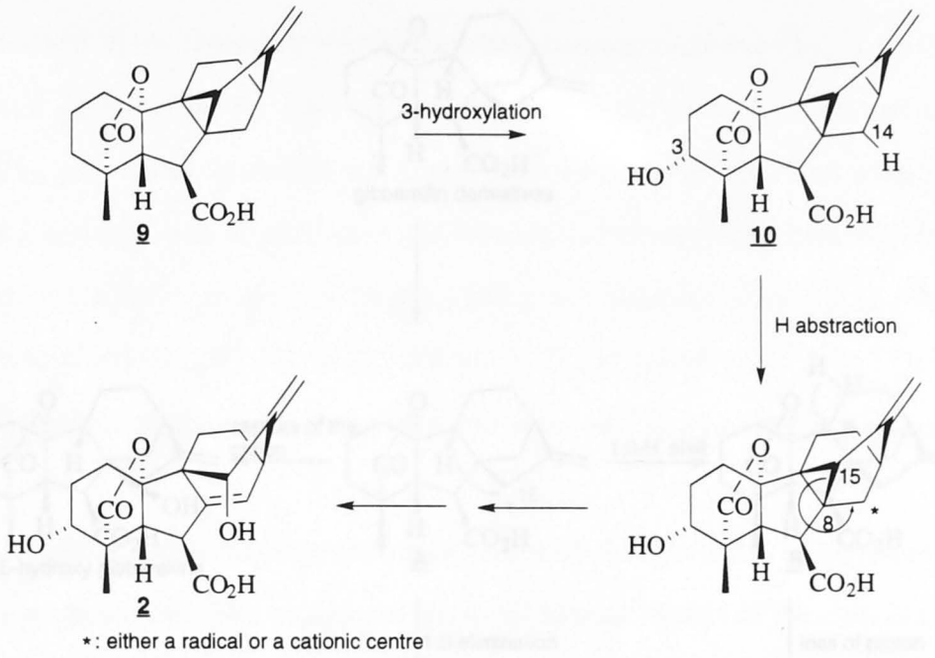


Figure 7. Mechanistic suggestion for the final stage of the biosynthesis of 2

Following on from these results, which show that pathway c in Figure 4 may be discarded, and the recent identification[92] of a 15-hydroxy gibberellin derivative as a natural antheridiogen in *A. Phyllitidis*, Mander[22] postulated the revised Figure 8. It seems that the biosynthesis may be commenced by the formation of a cationic centre at C(15) in a gibberellin molecule (cation **A**) *via* the action of an enzyme. Immediate capture of the cation by a source of nucleophilic OH would produce 15-hydroxy derivatives, while a 1,3-hydride transfer would give the more stable tertiary cation **B**. This pathway, consistent with pathway a in Figure 4 would be terminated by the loss of a proton from C(11) giving rise to GA_{73}-type compounds. A 1,3-elimination process would lead to the formation of 9,15-cyclo derivatives which would, *inter alia*, serve as precursors for the antheridane type of molecules.

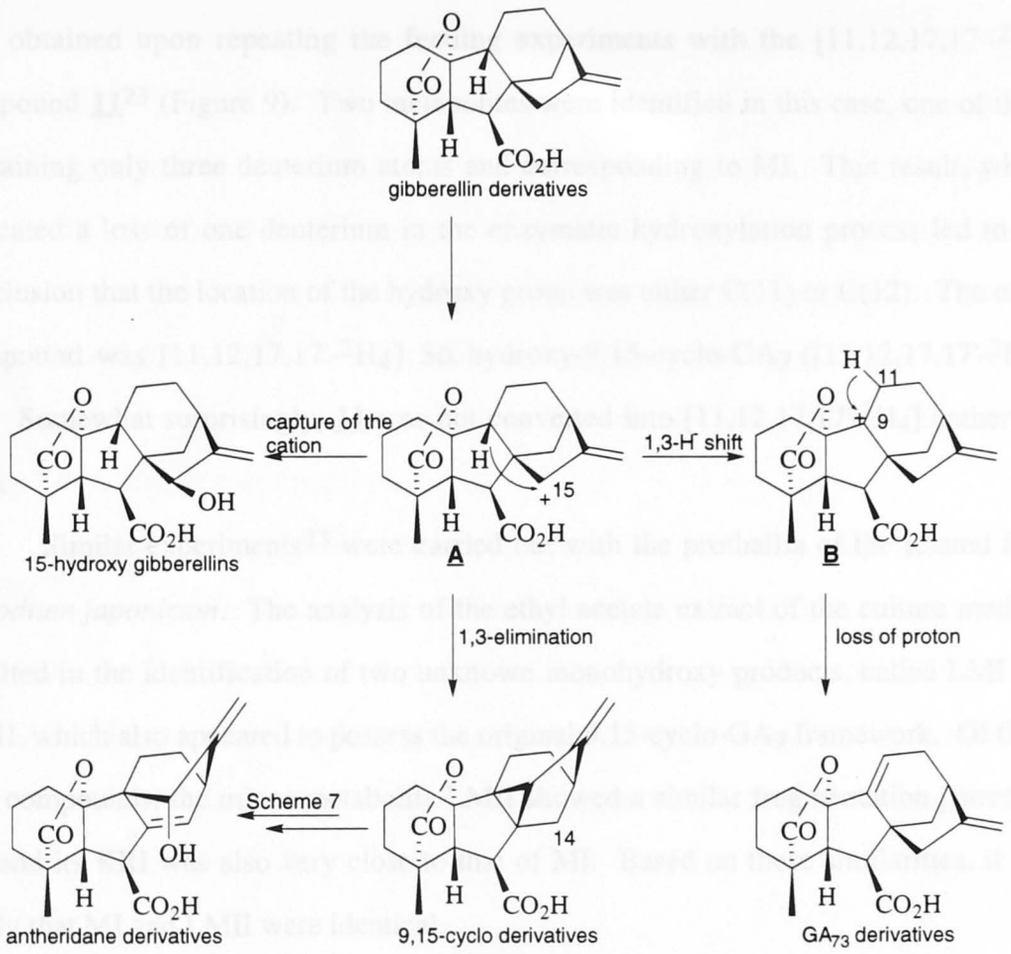


Figure 8. Revised version of the biosynthetic proposal

It is always possible that the conversion of [17,17'-$^2H_2$] 9,15-cyclo-GA_9 **7** into the *Anemia mexicana* antheridiogen ([17,17'-$^2H_2$]-**3**) observed during the biosynthetic experiments[21] might be due to non-specific metabolism because of a high dose of deuterium-labelled substrate. GC-MS analysis of the two unknown [$^2H_2$] monohydroxy compounds, which were named MI and MII revealed that they corresponded to the 9,15-cyclo-GA_9 type. Further information regarding the location of the hydroxy groups

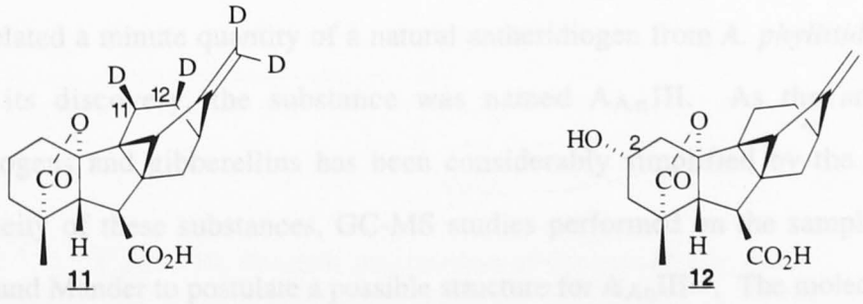


Figure 9. Structures of compounds 11 and 12

was obtained upon repeating the feeding experiments with the [11,12,17,17'-$^2H_4$] compound **11**[23] (Figure 9). Two metabolites were identified in this case, one of them containing only three deuterium atoms and corresponding to MI. This result, which indicated a loss of one deuterium in the enzymatic hydroxylation process led to the conclusion that the location of the hydroxy group was either C(11) or C(12). The other compound was [11,12,17,17'-$^2H_4$] 3α-hydroxy-9,15-cyclo-GA_9 ([11,12,17,17'-$^2H_4$]-**10**). Somewhat surprisingly, **11** was not converted into [11,12,17,17'-$^2H_4$] antheridic acid.

Similar experiments[23] were carried out with the prothallia of the related fern, *Lygodium japonicum*. The analysis of the ethyl acetate extract of the culture medium resulted in the identification of two unknown monohydroxy products, called LMI and LMII, which also appeared to possess the original 9,15-cyclo-GA_9 framework. Of these two compounds, the minor metabolite LMII showed a similar fragmentation pattern to MI and its KRI was also very close to that of MI. Based on these similarities, it was likely that MI and LMII were identical.

Metabolites MII and LMI matched each other both in terms of the mass spectrum fragmentation pattern and KRIs. Independent partial synthesis[24] later revealed that this compound corresponded to 2α-hydroxy-9,15-cyclo-GA_9 (**12**).

1.1.6 Isolation of a putative 3,12-dihydroxy-9,15-cyclo-GA_9 derivative

Prior to the biosynthetic studies described in the previous Section, Yamane's group isolated a minute quantity of a natural antheridiogen from *A. phyllitidis*. At the time of its discovery, the substance was named $A_{An}III$. As the analysis of antheridiogens and gibberellins has been considerably simplified by the structural homogeneity of these substances, GC-MS studies performed on the sample enabled Yamane and Mander to postulate a possible structure for $A_{An}III$[25]. The molecular mass of 504 (after pre-treatment with CH_2N_2 and BSTFA) corresponded to a regular C_{19} gibberellin with two hydroxy groups and an additional double bond or ring. The

prominent peak at *m/z* 129, characteristic of a 3-hydroxy group in a saturated A-ring (loss of C(1)-C(2)-C(3)-OTMS)[20,26], indicated that the other hydroxy group and the suspected ene function or ring were located in the C/D ring region. The mass spectrum was not consistent with the GA_{73} type[20,26] (such as **4**) and it did not show a loss of *m/z* 28 from the molecular peak, arising from the retro Diels-Alder reaction typical of the antheridane type[18] (such as **2**). Thus, it appeared that the additional degree of unsaturation may have been accounted for by the presence of the three-membered ring in the 9,15-cyclo type (such as **3**).

Another distinct loss which was observed, M^+-103, usually occurs in 12,13-dihydroxy GAs and corresponds to loss of CH_2OTMS[20,26]. Although this fragmentation is absent in the mass spectra of simple 12-hydroxy GAs[20,26], it was considered that the second hydroxy group was nevertheless most likely located at C(12) and that this particular mode of fragmentation was promoted by the strain caused by the cyclopropyl ring (Figure 10).

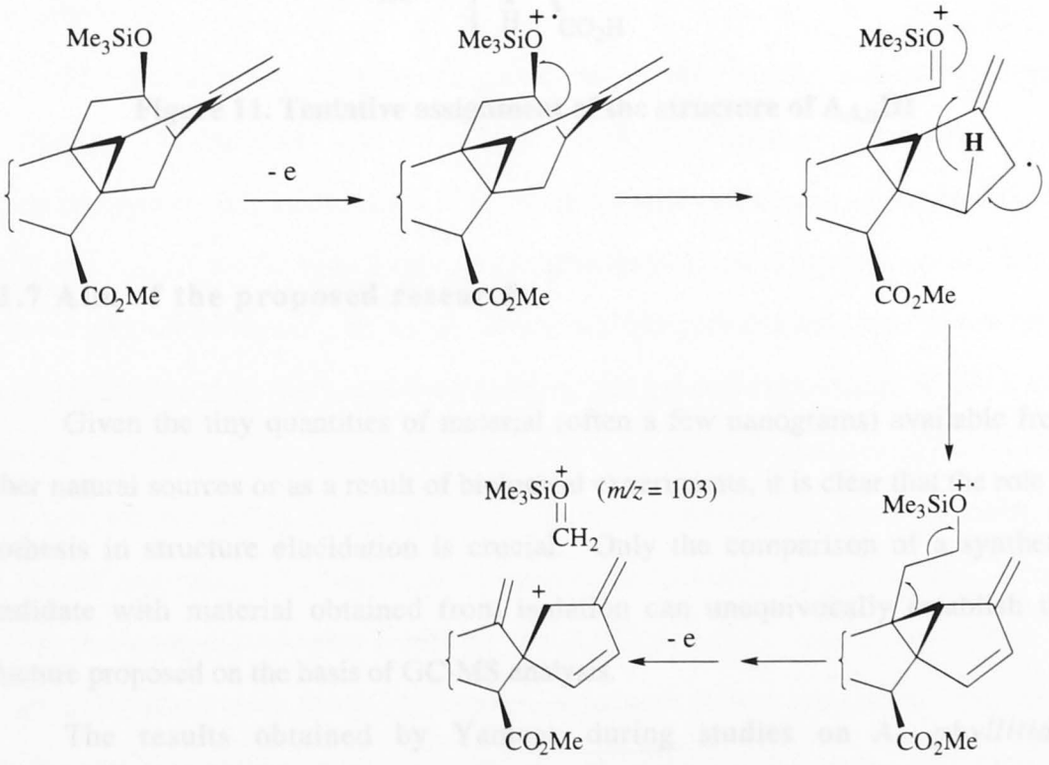


Figure 10. Possible mechanism of the loss of *m/z* 103

While 12-hydroxy GAs are widely distributed throughout the plant Kingdom, no 14-hydroxy compound has been isolated as yet[20]. The possibility of the hydroxy group

being attached to C(11) or C(13) could not be entirely excluded, but seemed to be rather remote. 3,11-Dihydroxy-9,15-cyclo-GA_9 methyl ester was later synthesized by Furber and Mander[27] (see Section 2.1, Chapter 2) and could be safely ruled out as a potential candidate. On the other hand, biosynthetic studies[21] on the 9,15-cyclo-GA_9 precursor **7**, which identified 3α-hydroxy-9,15-cyclo-GA_9 (**10**) as a natural antheridiogen in *A. phyllitidis*, and which demonstrated the ability of the enzymes of *A. phyllitidis* to hydroxylate the precursor at either C(11) or C(12), were supportive of the possibility that the C/D ring hydroxy group in $A_{An}III$ was located at C(12).

$A_{An}III$ was thus tentatively assigned as 3,12-dihydroxy-9,15-cyclo-GA_9 (Figure 11), on the assumption that the compound possessed the 9,15-cyclo framework[25].

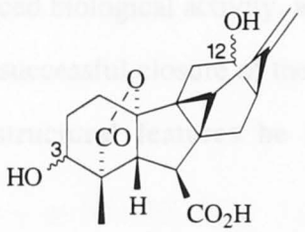


Figure 11. Tentative assignment of the structure of $A_{An}III$

1.1.7 Aim of the proposed research

Given the tiny quantities of material (often a few nanograms) available from either natural sources or as a result of biological experiments, it is clear that the role of synthesis in structure elucidation is crucial. Only the comparison of a synthetic candidate with material obtained from isolation can unequivocally establish the structure proposed on the basis of GC-MS analysis.

The results obtained by Yamane during studies on *A. phyllitidis* antheridiogens[21] shed further light on the biosynthesis of antheridic acid and disclosed another hydroxylation pathway in the processing of precursor **9** by *A. phyllitidis* enzymes. In addition, a new antheridiogen, the structure of which seemed to be related

to compounds identified during the biosynthetic studies and which might therefore be synthesized from the same precursor by identical or similar enzymes, was discovered[25]. It should be stressed though, that given the current state of knowledge about the proteins playing a role in *A. phyllitidis* pheromone synthesis, this possibility amounted to mere speculation.

Elucidating the structures of MI, LMII and A_{An}III would mean having to address the task of preparation of as many epimers of 11-hydroxy-, 12-hydroxy- and 3,12-dihydroxy-9,15-cyclo-GA_9 as possible, because GC-MS analysis does not provide definitive information about the relative configuration of stereocenters.

The partial synthesis of 11-hydroxy and 12-hydroxy gibberellins poses a major challenge to a synthetic chemist. The 12-hydroxy compounds, which are increasingly in demand, due to their pronounced biological activity, are among the least accessible of the natural gibberellins[20]. The successful closure of the 9,15-cyclopropyl ring requires that certain strictly defined structural features be introduced into a gibberellin molecule[28,29].

The primary aim of research described in this Thesis was to synthesize 11-hydroxy-, 12-hydroxy- and 3,12-dihydroxy-9,15-cyclo-GA_9 derivatives from commercially available gibberellins. The comparison of synthetic compounds with MI, LMII and A_{An}III would help establish the structures of these substances and hence improve our understanding of the *A. phyllitidis* biosynthetic enzymes involved in pheromone production. Although the possibility that synthetic derivatives would not be matched with these compounds had to be admitted, given the limitations of the GC-MS technique, the probability of their discovery in future investigations was high, based on the natural occurrence of a number of 11-hydroxy and 12-hydroxy gibberellins. In this regard, the synthetic samples would be a significant contribution to the library of compounds, which serve for GC-MS comparisons with natural substances. Exploring possible synthetic pathways might establish a reliable approach to this rare class of compounds, the 3-hydroxy series in particular, and lead to the preparation of intermediates from which other natural antheridiogens and gibberellins could be derived.

1.2 GENERAL CONSIDERATIONS FOR C-RING FUNCTIONALISATION COMBINED WITH THE 9,15-RING CLOSURE

The density of functionality on the highly strained skeleton of gibberellins has ensured a fascinating variety of chemical reactions. The chemistry of gibberellins, which has been developed over the past few decades, has been thoroughly reviewed recently[20,30]. The aim of this Section is to give a brief survey of several reactions and strategies with a view to providing a background to the synthetic endeavours attempted during this work. Some of them are described or analyzed in detail in the Introductions to separate Chapters, illustrating the considerations which led to the design of a particular synthetic pathway.

1.2.1 Starting materials

Two highly functionalized derivatives, GA_3 (**13**) and GA_7 (**14**) (Figure 12), the structural features of which are favourable for further functional group manipulation, are commercially available at present[20]. **13** is produced more efficiently than **14** and is therefore less expensive, while the latter compound is obtained as a mixture with its 1,2-dihydro counterpart, GA_4 (cf. Section 2.3.1, Chapter 2).

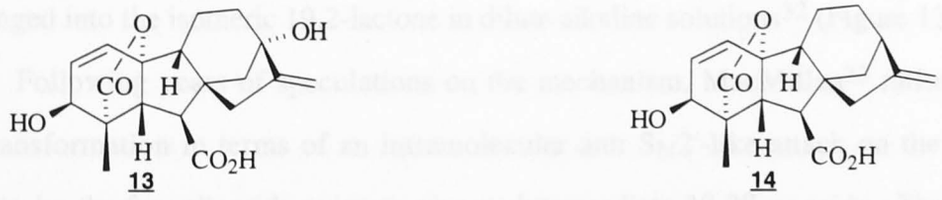


Figure 12. Structures of commercially available gibberellins

1.2.2 Classification of strategies

Given the structures of the starting materials and the wide array of reactions which are used to manipulate gibberellin molecules, there appear to be three basic strategies for the insertion of functional groups into the C-ring of gibberellins[31]:

1. Extending the A-ring functionality.
2. Effecting a transannular process from substituents attached to the D-ring.
3. Cleaving the C(13)-C(16) bond and rebuilding the D-ring at a later stage.

Pursuing any of these three options, it should be possible to introduce either the desired hydroxy group or any other synthetic function, which may be elaborated towards this goal, such as a double bond or a suitable ligand, which may be either selectively replaced or modified. The introduction of the cyclopropyl ring described in detail in Section 1.2.4 may be classified as a type 1 strategy.

1.2.3 Type 1 strategies

The more successful strategies using the A-ring functionality in GA_3 or GA_7 type of molecules to introduce synthetic functions into the C-ring are, by and large, based on 19,10 → 19,2 lactone isomerizations and related reactions (*vide infra*). It has been known since the early 1960s that the allylic lactone moiety in GA_3 (**13**) can be rearranged into the isomeric 19,2-lactone in dilute alkaline solutions[32] (Figure 13).

Following years of speculations on the mechanism, MacMillan[33] rationalized this transformation in terms of an intramolecular anti S_N2'-like attack on the allylic function by the free alkoxide anion to give an intermediate 2β,3β-epoxide. The epoxy function is subsequently cleaved by an S_N2-like opening by the carboxylate group.

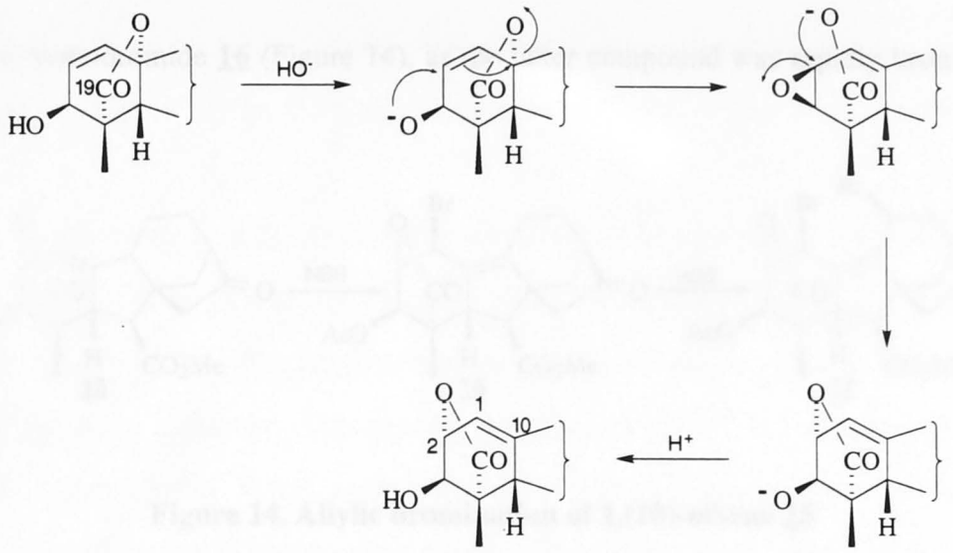


Figure 13. Mechanism of HO⁻-mediated 19,10 → 19,2 A-ring lactone reversion

The driving force for the overall process seems to be provided by the relief of the ring strain in going from the trans A/B ring junction in the starting 19,10-lactone to the 1,10-double bond in the 19,2-lactone[33]. Equivalent processes have also been observed for GA_7 (**14**)[29]. The isomerization may also be effected by palladium acetate and Pd^0 complexes[34], or weak Lewis acids, such as ferric chloride[35] and zinc bromide[29]. Although no attempt has been made to explain the mechanisms of either Pd or Lewis acid-induced reversions of the lactone function, it can well be assumed that η^3-complexes of Pd are involved in the former. These methods have been successfully employed on 3-acetates, while the hydroxide ion-mediated rearrangements require that the 3-OH group be unprotected.

The double bond in the 1,10 position renders C(9) allylic and, consequently, suitably selected allylic functionalization processes can extend the functionality in the C-ring direction. An efficient example of the synthetic use of these reactions can be demonstrated by the key step in the synthetic studies on 9,15-cyclo-GA_9 methyl ester (**9** methyl ester) and related compounds by Furber and Mander[29]. Allylic bromination of 1,(10)-olefin **15** (obtained by $ZnBr_2$-mediated allylic lactone reversion from its 1,2-counterpart) was envisaged to take place with the reaction of the initially formed allylic radical at the sterically less hindered C(1) position. In the event, the 1β,11β-dibromo compound **17** was obtained in an excellent yield from the initially

formed monobromide **16** (Figure 14), as the latter compound was rapidly brominated at C(11).

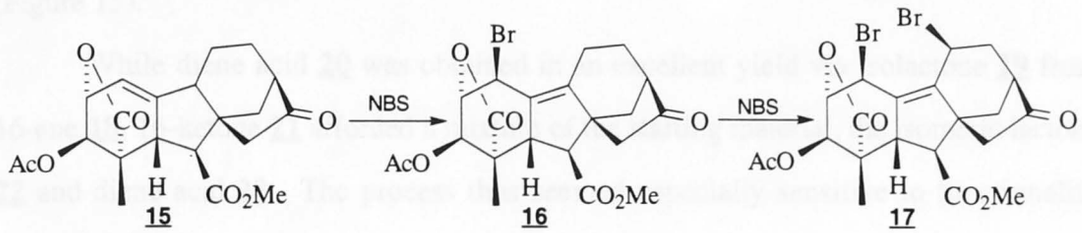


Figure 14. Allylic bromination of 1,(10)-alkene 15

This step set the stage for further transformations and introduced functionality into the C-ring. Compound **17** could thus serve as a key intermediate in the synthesis of 3,11-dihydroxy-9,15-cyclo-GA_9 methyl esters[27] (see Section 2.1, Chapter 2).

It has been shown that the 3-hydroxy group, either free or protected, has a major

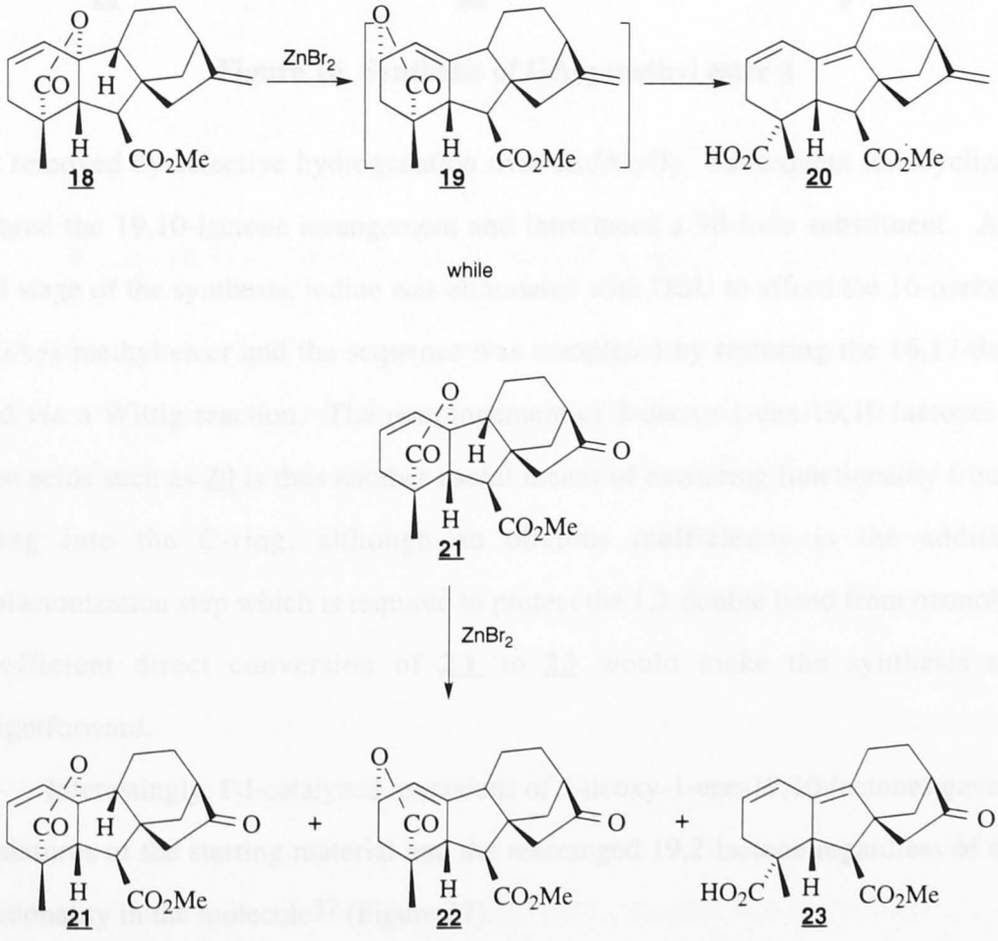


Figure 15. $ZnBr_2$-induced rearrangements of 3-deoxy compounds

influence on these reactions. When Lewis acid-induced rearrangements were attempted on 3-deoxy-1-enes of the GA_3 and GA_7 type, different results were obtained[36,37] (Figure 15).

While diene acid **20** was obtained in an excellent yield via isolactone **19** from 16-ene **18**, 16-ketone **21** afforded a mixture of the starting material, the isomeric lactone **22** and diene acid **23**. The process thus seemed especially sensitive to functionality elsewhere in the molecule. This reaction found application[36] in the synthesis of the natural antheridiogen, GA_{73} methyl ester (**4**). Thus, the triene compound **20** was converted into ketone **23** in three steps (Figure 16) and then the 1,2-double bond in **23**

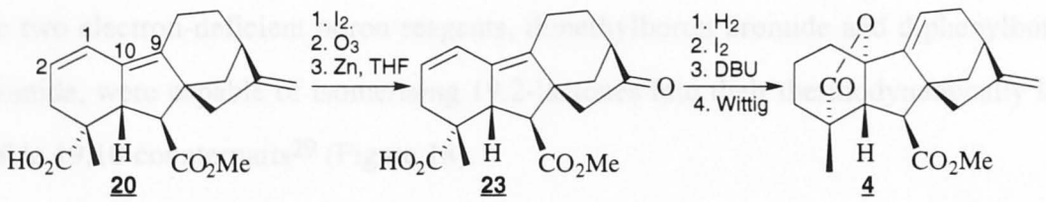


Figure 16. Synthesis of GA_{73} methyl ester 4

was removed by selective hydrogenation over Rh/Al_2O_3. Subsequent iodocyclization restored the 19,10-lactone arrangement and introduced a 9β-iodo substituent. At the final stage of the synthesis, iodine was eliminated with DBU to afford the 16-norketone of GA_{73} methyl ester and the sequence was completed by restoring the 16,17-double bond *via* a Wittig reaction. The rearrangement of 3-deoxy-1-ene-19,10-lactones into diene acids such as **20** is thus another useful means of extending functionality from the A-ring into the C-ring, although an obvious inefficiency is the additional iodolactonization step which is required to protect the 1,2-double bond from ozonolysis; an efficient direct conversion of **21** to **23** would make the synthesis more straightforward.

Interestingly, Pd-catalyzed reversions of 3-deoxy-1-ene-19,10-lactones gave rise to mixtures of the starting material and the rearranged 19,2-lactone regardless of other functionality in the molecule[37] (Figure 17).

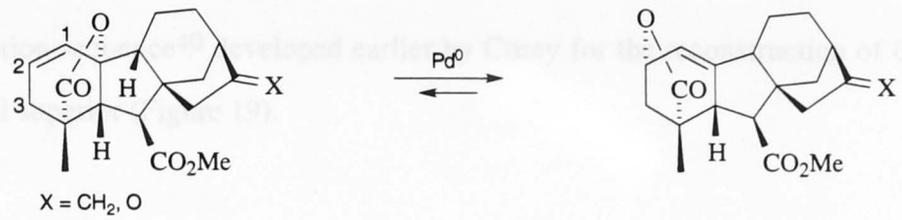


Figure 17. Pd^0-catalyzed A-ring lactone isomerizations

It was concluded by Kraft-Klaunzer and Mander[38] that in the cases where mixtures were obtained with Pd^0 and $ZnBr_2$, the reactions had reached equilibrium, but no mechanistic studies to substantiate this were undertaken.

A reverse process, i.e. 19,2 → 19,10 lactone rearrangement, which is general for a wide range of gibberellin structures, has also been discovered. It was found that the two electron-deficient boron reagents, dimethylboron bromide and diphenylboron bromide, were capable of isomerising 19,2-lactones into their thermodynamically less stable 19,10-counterparts[29] (Figure 18).

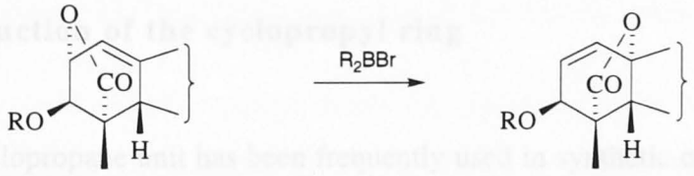


Figure 18. Boron-induced 19,2 → 19,10 A-ring lactone reversions

Mander explained this in terms of the coordination between the lactone function and the borane followed by an S_N2 substitution by bromide at C(2) and completed by an intramolecular S_N2'-like substitution at C(10) by the liberated carboxylate[29]. The contrathermodynamic nature of this process can be rationalized to some extent by the assumption that it is driven by selective complexation between the borane and the 19,2-lactone isomers and that the 19,10-lactone function is too sterically hindered to form an adduct. Although this reaction was successfully applied to 3-substituted compounds, complex mixtures of products were formed in preliminary experiments with 3-deoxy-1(10)-ene-19,2-lactone compounds[39]. This process provided the advantage of having the potential to rearrange a 19,2-lactone into its 19,10-congener in one step. For 3-deoxy compounds though, the hydrolysis/ iodolactonization/ reductive

elimination sequence[40] developed earlier by Corey for the reconstruction of GA_3 (**13**) was still superior (Figure 19).

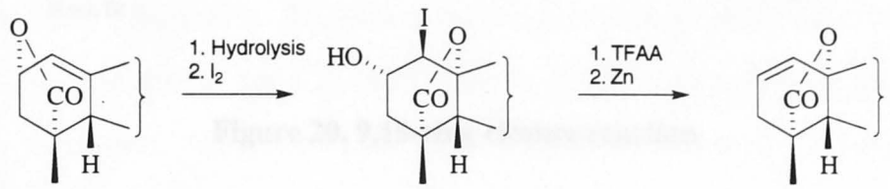


Figure 19. Corey's protocol applied to 19,2 → 19,10 A-ring lactone isomerisation

This Section can be concluded by noting that the A-ring lactone rearrangements open up pathways to the insertion of functionality into the C-ring and that they can be carried out in both directions. The drawback appears to be that the C-ring positions directly accessible in this way are mainly C(9) and C(11).

1.2.4 Introduction of the cyclopropyl ring

The cyclopropane unit has been frequently used in synthetic organic chemistry as a precursor of larger rings[41] or 1,3-disubstituted systems[41,42]. It may therefore be regarded as a unique type of functionality and in view of the reactions preceding the 9,15-ring closure, the introduction of the cyclopropyl ring can be formally classified as a type 1 procedure; the functionality was extended from the A-ring into the C-ring prior to the ring-forming reaction.

In order to form a bond between C(9) and C(15), a 16-keto-1-halo-9-ene arrangement was introduced into a gibberellin molecule. With the appropriate functionality in place, a base abstracts a proton from C(15) and the enolate undergoes intramolecular alkylation with the 1-halo-9-ene moiety[28,29] (Figure 20).

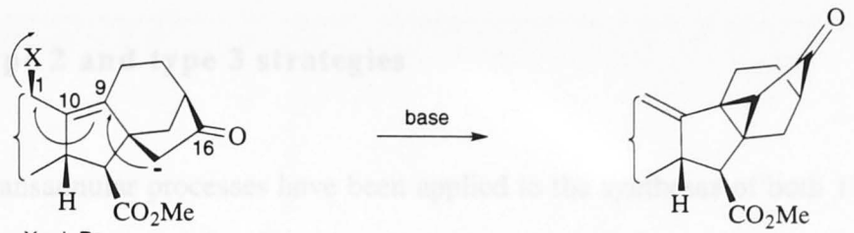


Figure 20. 9,15-ring closure reaction

The original route to 9-ene-1-halides was based on the reaction of GA_7 type derivatives, such as **24**, with hydrazine hydrate in DMF[29] (Figure 21).

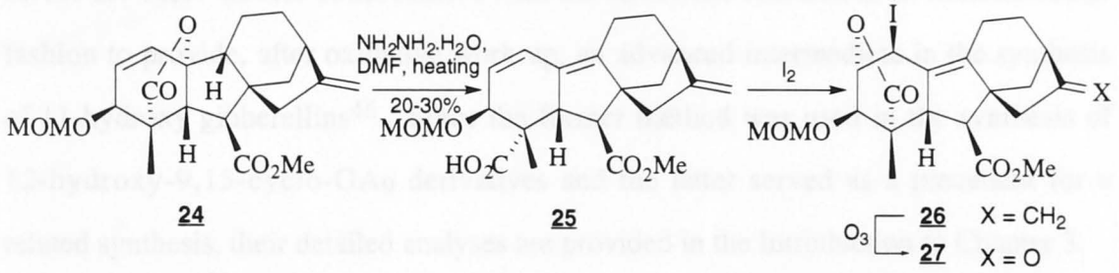


Figure 21. Original preparation of 9,10-ene-1-halides

The low yield of this reaction, however, created a major bottleneck in the accumulation of adequate supplies of intermediates corresponding to **25**. The iodocyclization of 1,9-diene acids of this type proceeds with the internal carboxy nucleophile attacking the less substituted 1,2-double bond after activation by the iodine electrophile. The reaction is controlled by stereoelectronic Fürst-Plattner effects[83a], thereby providing access to 1β-iodo lactones[16,36].

The reactions of 3-deoxy-1-ene-19,10-lactones described in the previous Section contributed a great deal to resolving the problem of efficient access to 1,9-diene acids (*vide supra*), providing a ready supply of these compounds, although the prerequisite for this method to be successfully applied appeared to be the absence of the 3-hydroxy function. It should also be noted that the allylic functionalization of 1,(10)-ene compound **15**[29] (Figure 4) introduced the crucial 9-ene-16-keto-1-halo arrangement as well.

1.2.5 Type 2 and type 3 strategies

Transannular processes have been applied to the syntheses of both 11-hydroxy and 12-hydroxy gibberellins. The idea of this concept is based on the functionalisation of the exocyclic double bond in the D-ring to afford a methylene group with an auxiliary substituent, $-CH_2R$. In cases where R = OH, the remote site functionalisation reaction with $Pb(OAc)_4$ (later $Ph(IOAc)_2$) was employed to introduce a hydroxy group at C(12)[43,44,45]. The C(17)-BH_2 function formed by the addition of a borane reagent across the 16,17-double bond, reacted with the 9,(11)-ene function in an intramolecular fashion to provide, after oxidative work-up, an advanced intermediate in the synthesis of 11-hydroxy gibberellins[46]. Since the former method was used in the synthesis of 12-hydroxy-9,15-cyclo-GA_9 derivatives and the latter served as a precedent for a related synthesis, their detailed analyses are provided in the Introduction to Chapter 3.

The type 3 methods do not seem to have found a significant application in the partial synthesis of C-ring functionalized gibberellins as yet.

CHAPTER 2

2. SYNTHESIS OF 11-HYDROXY-9,15-CYCLO-GA$_9$ METHYL ESTERS

2.1 INTRODUCTION

As mentioned in Chapter 1, the task of synthesizing 11-hydroxy-9,15-cyclogibberellins has already been addressed by Mander and Furber[27]. They prepared 3β,11α-dihydroxy-9,15-cyclo-GA$_9$ **33** and its 3α–epimer **34** by means of an efficient, 10-step synthesis (Figure 22) from GA$_7$ methyl ester **28**.

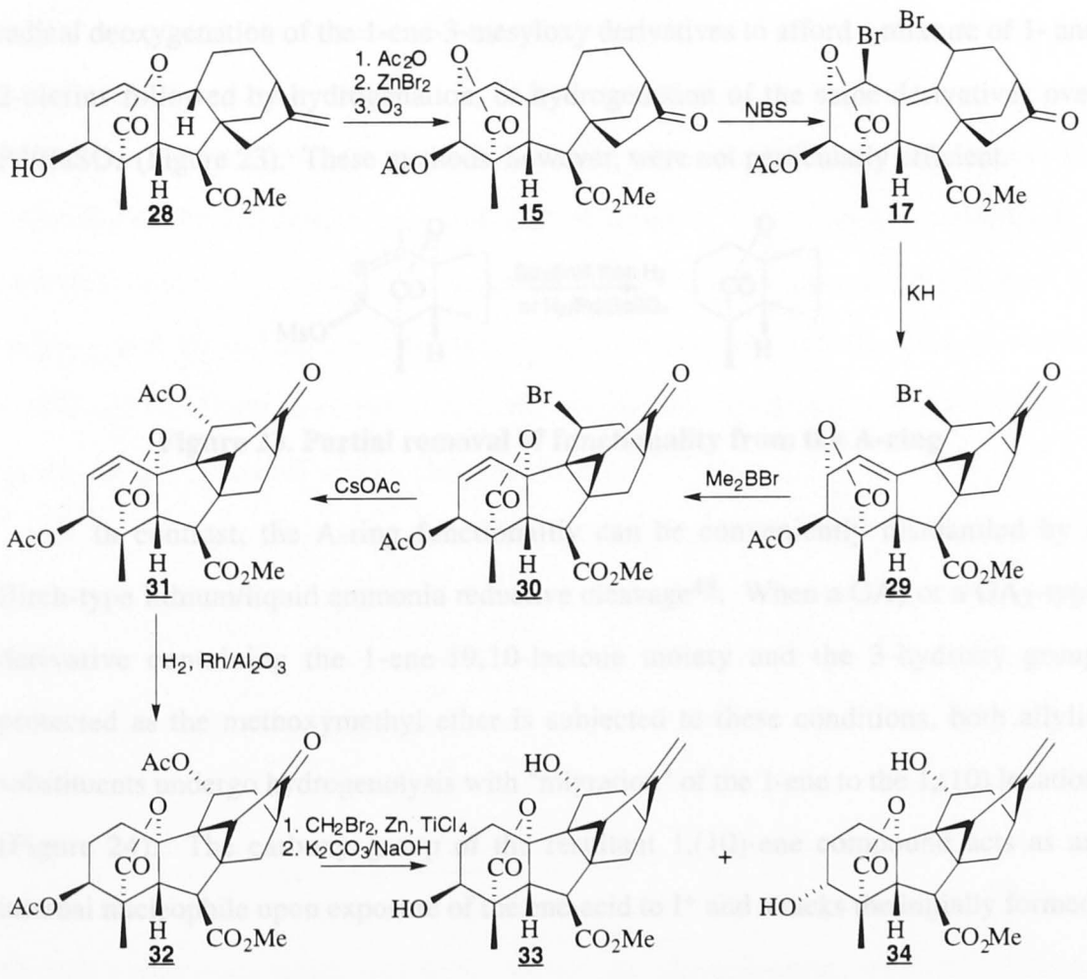


Figure 22. Synthesis of 3,11-dihydroxy-9,15-cyclo-GA$_9$ methyl esters

The key step of the sequence was the allylic bromination of ketone **15**, which was originally designed to introduce the 9-ene-1-halo moiety required for the 9,15-ring closure. In the event, the bromination not only achieved this goal, but also inserted another β-bromo substituent at C(11) to give dibromide **17** (Figure 14, Chapter 1). With this functionality in place, the cyclopropyl ring was closed with potassium hydride and the synthesis could be completed by converting the 9,15-cyclo derivative **29** into the target compounds in a straightforward series of steps.

Following this study, it would seem obvious that the 3-deoxy series of 11-hydroxy-9,15-cyclogibberellins could be prepared by partial removal of functionality from the A-ring at the stage of compound **31**. The methodology[29,47] which had been used so far to remove the 1-ene-3-hydroxy arrangement involved either radical deoxygenation of the 1-ene-3-mesyloxy derivatives to afford a mixture of 1- and 2-olefins followed by hydrogenation, or hydrogenation of the same derivatives over $Pd/BaSO_4$ (Figure 23). These methods, however, were not particularly efficient.

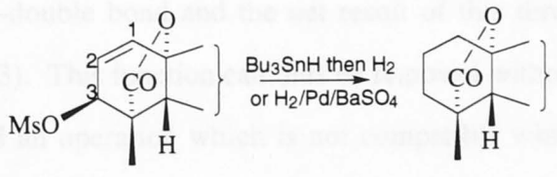


Figure 23. Partial removal of functionality from the A-ring

In contrast, the A-ring functionality can be conveniently dismantled by a Birch-type lithium/liquid ammonia reductive cleavage[48]. When a GA_7 or a GA_3-type derivative containing the 1-ene-19,10-lactone moiety and the 3-hydroxy group protected as the methoxymethyl ether is subjected to these conditions, both allylic substituents undergo hydrogenolysis with "migration" of the 1-ene to the 1,(10) location (Figure 24). The carboxy group of the resultant 1,(10)-ene compound acts as an internal nucleophile upon exposure of the ene-acid to I^+ and attacks the initially formed

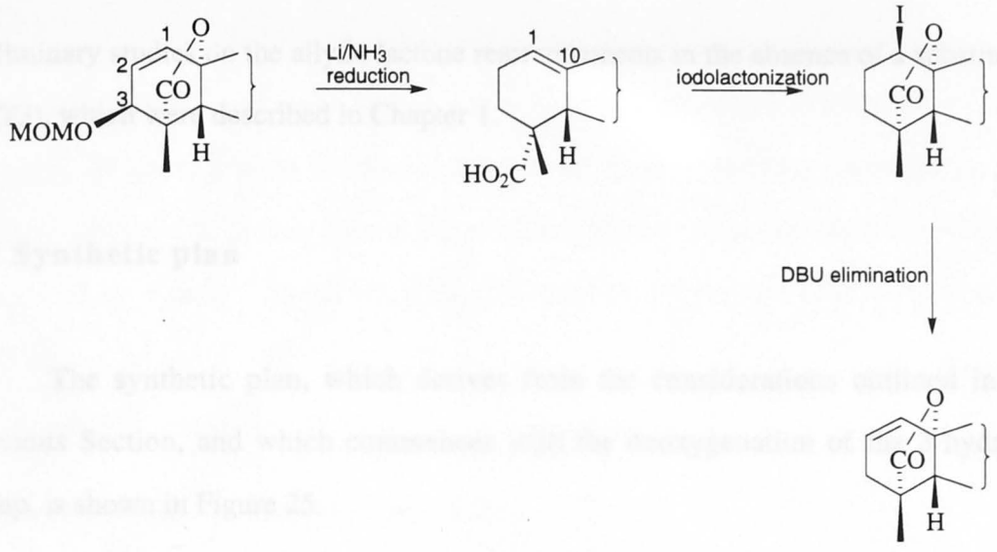


Figure 24. Formal deoxygenation of the 3-hydroxy group by a Birch-type process

β-face iodonium ion in accordance with both electronic and stereoelectronic factors at C(10) thereby producing a 1β-iodo-19,10-lactone, the 1β-iodo substituent of which may be used as a precursor of the 1-ene function[36]. A DBU-induced elimination reintroduces the 1,2-double bond and the net result of this three-step sequence is a deoxygenation of C(3). This function can thus be removed without the use of toxic tin reagents and, should an operation which is not compatible with the presence of the 1,2-double bond have to be performed (such as the ozonolysis of the 16-ene moiety), it can be carried out on the intermediate iodolactone[37].

In principle, this sequence could be employed for the partial removal of functionality from the A-ring in the synthesis of the 3-deoxy series of the 11-hydroxy-9,15-cyclo compounds. As compared to the original route of Furber[27], the sensitivity of the cyclopropyl ring towards the conditions of the alkali metal/ammonia reduction[41,49,50] changes the timing of the operations to be undertaken, as the Birch-type deoxygenation must be carried out first.

The objective of work described in this Chapter was to develop a synthesis of the title compounds which would incorporate the aforementioned alkali metal-based method for the removal of the 3-hydroxy group as the first stage. Apart from avoiding the problems associated with the methods for the partial removal of the A-ring functionality outlined in Figure 23, it would provide the opportunity to continue the

preliminary studies on the allylic lactone rearrangements in the absence of a substituent at C(3), which were described in Chapter 1.

2.2 Synthetic plan

The synthetic plan, which derives from the considerations outlined in the previous Section, and which commences with the deoxygenation of the 3-hydroxy group, is shown in Figure 25.

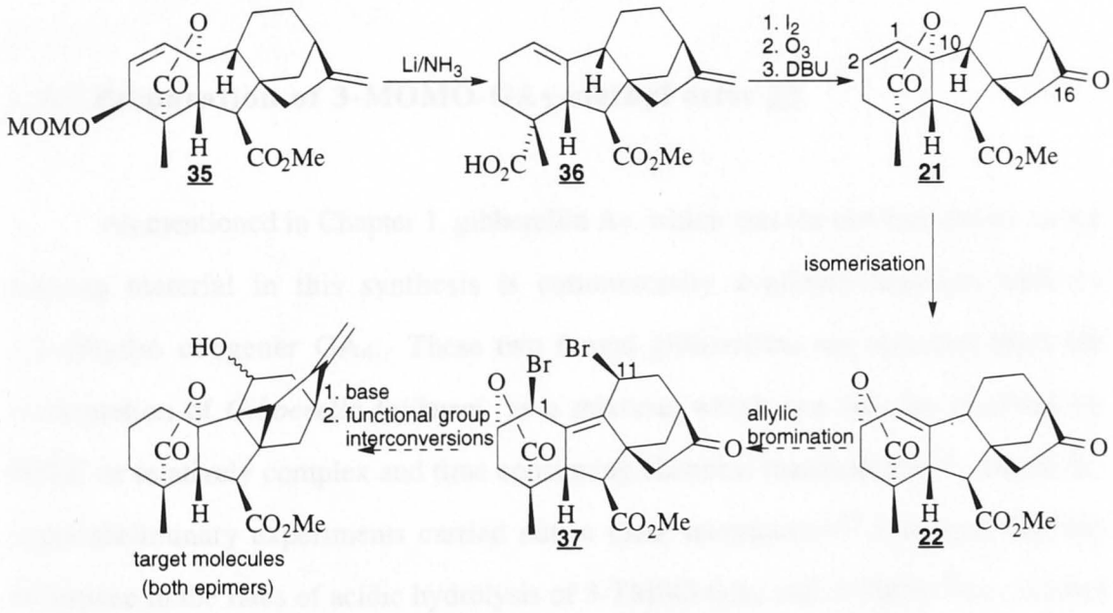


Figure 25. Synthetic plan

The treatment of the starting material, 3-MOMO-GA_7 methyl ester **35** with lithium in liquid ammonia was known[36] to afford the 1,(10)-ene acid **36** and, with this compound in hand, the iodolactonization/DBU elimination sequence could be performed in order to restore the 1-ene function[36,37]. As the goal of the following procedures would be the introduction of the 9-ene-1-halo-16-oxo arrangement, the ozonolysis step could be inserted between the iodolactonization and elimination steps in order to replace the 16,17-double bond with the oxo function[37]. The resultant 1-ene-16-oxo-19,10-lactone compound **21** could be isomerised into its 19,2-lactone counterpart **22** *via* a palladium-based procedure[37] and this derivative could subsequently be subjected to allylic bromination. Similar to the conversion of **15** to

17[29], this process could be expected to deliver the 1β,11β-dibromide **37**, containing the required synthetic functions [1-bromo-9-ene-16-oxo moiety and a substituent at C(11)] in place. **37** could thus be elaborated towards the target molecules commencing with the base-mediated 9,15-ring closure (Figure 20, Chapter 1) and the synthesis would be completed by standard functional group interconversion reactions, as in the route of Furber[27].

2.3 PREPARATION OF STARTING MATERIAL

2.3.1 Preparation of 3-MOMO-GA_7 methyl ester 35

As mentioned in Chapter 1, gibberellin A_7, which was the obvious choice as the starting material in this synthesis is commercially available together with its 1,2-dihydro congener GA_4. These two fungal gibberellins are obtained from the fermentation of *Gibberella fujikuroi* as a mixture, which can only be resolved by HPLC or relatively complex and time consuming chemical manipulation[51]. However, some preliminary experiments carried out in these laboratories[52] indicated that the difference in the rates of acidic hydrolysis of 3-TMSO-GA_4 and 3-TMSO-GA_7 methyl esters was large enough to discriminate between them. In order to generate a ready supply of 3-MOMO-GA_7 methyl ester, the commercially available mixture of gibberellins A_4 (**38**) and A_7 (**14**) (Figure 26) was sequentially treated with MeI/K_2CO_3, to esterify the 7-carboxy group, and trimethylsilyl chloride to silylate the 3-hydroxy functions. Upon treatment with acetic acid in methanol, **40** was hydrolysed faster than **39** thereby furnishing GA_7 methyl ester (**28**), which was easily separated by column chromatography. The hydrolysis could be conveniently monitored by TLC based on the different colour reactions of GA_7 and GA_4 derivatives upon visualization with vanillin in sulfuric acid (see Experimental, page 120). A more precise ^1H NMR analysis showed that **28** was obtained in a 90:10 mixture with GA_4 methyl ester (formed by slow hydrolysis of 3-TMSO-GA_4 methyl ester **39**), with the actual amount

being dependent on the composition of the particular batch of GA_4/GA_7. Further purification of this material for synthetic purposes was not necessary. The difference in the rates of hydrolysis is presumably due to the pseudoaxial 3-OTMS group in **40**, which is located in a slightly less sterically crowded environment than the axial 3-OTMS group in **39**, thus being more susceptible to acid hydrolysis[53a]. This methodology provided easy access to **28** from the commercial source without recourse to extensive chemical operations.

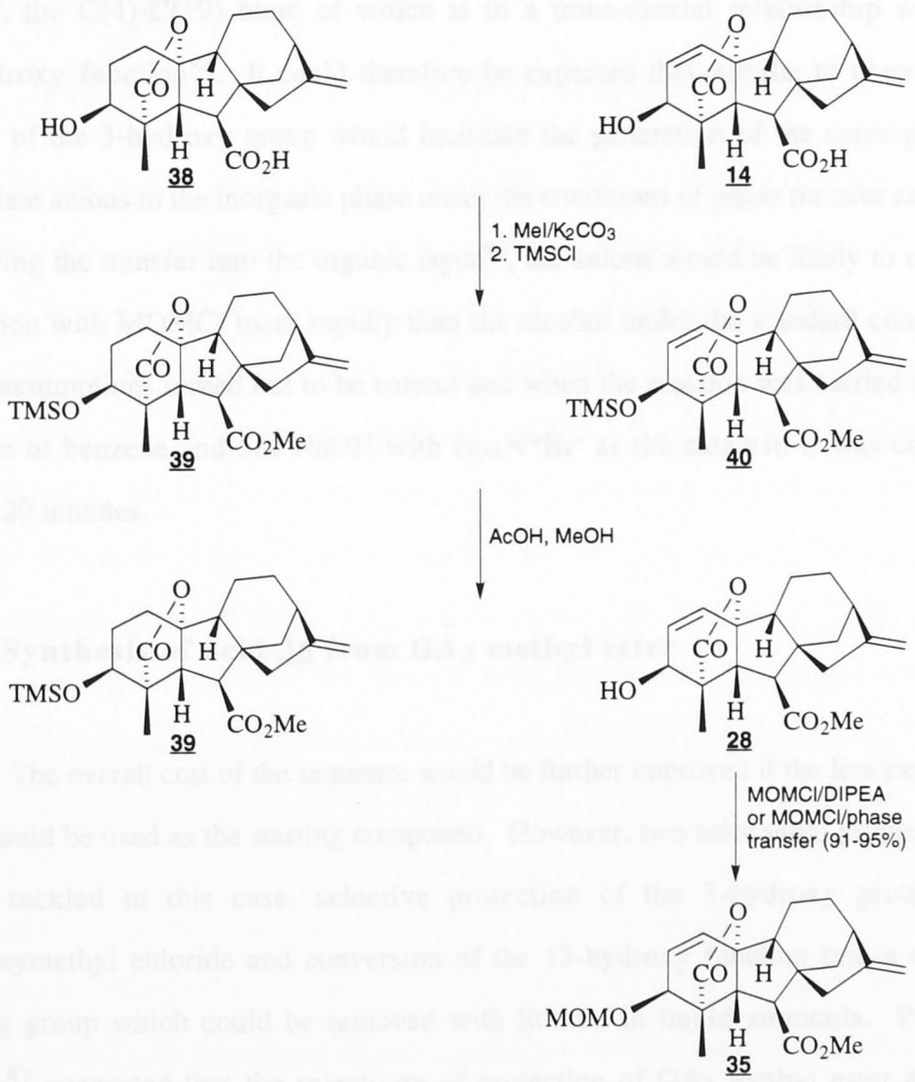


Figure 26. Preparation of 3-MOMO-GA_7 methyl ester

Having established the method for obtaining GA_7 methyl ester from the GA_4/GA_7 mixture, the sequence could be commenced by the derivatization of **28** with methoxymethyl chloride. The reaction of the 3β-hydroxy group of gibberellins with

MOMCl is usually a slow process under the standard conditions, involving treatment of a 3β-hydroxy gibberellin derivative with this reagent in the presence of diisopropylethyl amine and dimethylaminopyridine[29]. It has been demonstrated[56] that phenols, the hydroxy groups of which are more acidic than those of aliphatic alcohols, can be efficiently converted into their MOM-ethers under phase transfer conditions. It could be speculated that the 3β-hydroxy group of gibberellins is probably more acidic than an ordinary secondary hydroxy function, owing to the electronic effect of the 19-carbonyl moiety, the C(4)-C(19) bond of which is in a trans-diaxial relationship with the 3β-hydroxy function[54]. It could therefore be expected that, similar to phenols, the acidity of the 3-hydroxy group would facilitate the generation of the corresponding alcoholate anions in the inorganic phase under the conditions of phase transfer catalysis. Following the transfer into the organic layer[55], the anions would be likely to undergo alkylation with MOMCl more rapidly than the alcohol under the standard conditions. These assumptions turned out to be correct and when the reaction was carried out in a mixture of benzene and 5M NaOH with $Bu_4N^+Br^-$ as the catalyst, it was complete within 20 minutes.

2.3.2 Synthesis of acid 36 from GA_3 methyl ester

The overall cost of the sequence would be further improved if the less expensive GA_3 could be used as the starting compound. However, two substantial problems had to be tackled in this case: selective protection of the 3-hydroxy group with methoxymethyl chloride and conversion of the 13-hydroxy function into a suitable leaving group which could be removed with lithium in liquid ammonia. Previous results[57] suggested that the selectivity of protection of GA_3 methyl ester **45** with MOMCl under the standard conditions (CH_2Cl_2, DIPEA, DMAP) was very low. It was also reported that the 13-acetoxy group was simply hydrolyzed[58] when the 3-MOMO-13-AcO-GA_3 methyl ester **41** was subjected to lithium/ammonia reduction, while the same 13-acetoxy moiety was reductively cleaved[59] from a slightly different gibberellin derivative **43** under the same conditions (Figure 27).

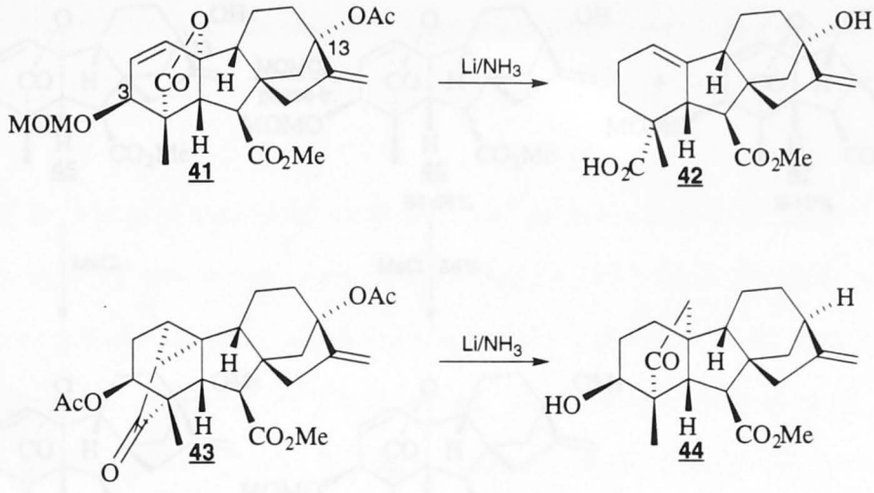


Figure 27. Cleavage of the 13-AcO group in Li/NH_3 reductions

Similar to the example of GA_7 methyl ester described in the previous Section, the phase transfer procedure utilizing the increased acidity of the 3-hydroxy group appeared to provide the opportunity for the discrimination between the 3- and 13-hydroxy functions in the alkylation reaction with methoxymethyl chloride. It could be expected that the alcoholate anion would be formed predominantly from the more acidic[54] and less hindered 3-hydroxy function and, following the alkylation of the anion with MOMCl in the organic phase, the solubility of the resulting 3-MOMO derivative in the inorganic phase would be significantly diminished. Indeed, treatment of GA_3 methyl ester (**45**) with MOMCl in a two-phase medium of benzene/ EtOAc/ 5M NaOH with tetrabutylammonium bromide as the catalyst afforded a mixture of 3-MOMO and 3,13-di-MOMO derivatives in the ratio of 10:1 (Figure 28). With the A-ring functionality in place for the reductive cleavage process, the 3-MOMO compound **46** was converted into the 3-MOMO-13-MsO-GA_3 methyl ester (**49**), as the 13-mesyloxy group was not likely to be hydrolyzed under the reaction conditions and it was superior to the acetoxy group in terms of its leaving ability. As expected, when compound **49** was treated with lithium in liquid ammonia, the desired derivative **36** was obtained in a good yield.

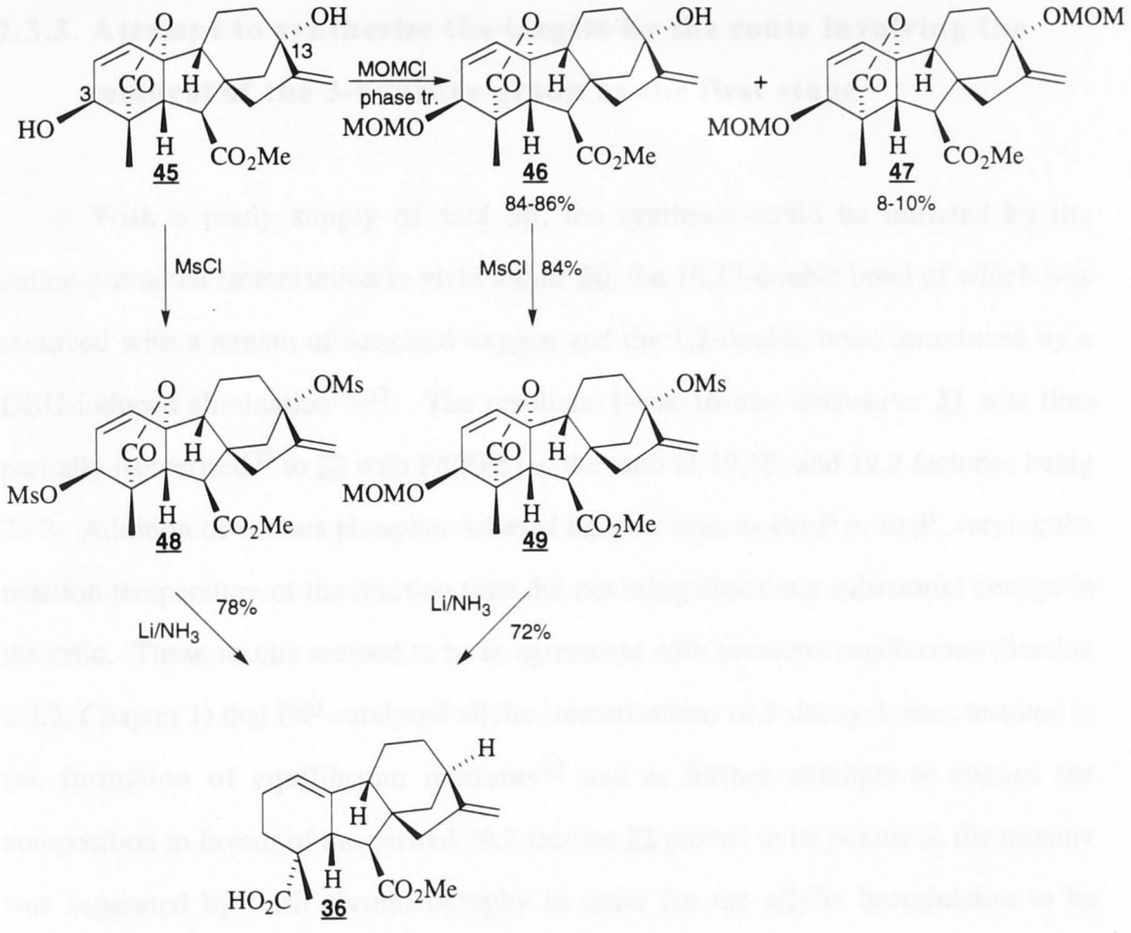


Figure 28. Synthesis of acid 36

Finally, in an attempt to shorten the route further and because the phase transfer method turned out to be less efficient on a scale larger than 100 mg (furnishing considerable amounts of byproducts), GA_3 methyl ester was treated directly with mesyl chloride to afford the 3,13-di-MsO compound **48**[97]. It could be reasoned that if the methoxymethyloxy group with its poor leaving ability was reductively removed with alkali metal in liquid ammonia, so would be its 3-mesyloxy counterpart. Accordingly, when the dimesylate **48** was exposed to these conditions, the acid **36** was obtained as the sole product, the spectral data of which were fully in accord with those previously reported for this compound.

2.3.3. Attempt to synthesize the targets by the route involving the removal of the 3-hydroxy group as the first stage

With a ready supply of acid **36**, the synthesis could be initiated by the iodine-promoted lactonisation to yield iodide **50**, the 16,17-double bond of which was removed with a stream of ozonised oxygen and the 1,2-double bond introduced by a DBU-induced elimination[36,37]. The resultant 1-ene-16-oxo derivative **21** was then partially isomerised[37] to **22** with $Pd(PPh_3)_4$, the ratio of 19,10- and 19,2-lactones being 2 : 3. Addition of various phosphorus-based ligands, such as Bu_3P or Et_3P, varying the reaction temperature or the reaction time did not bring about any substantial change in the ratio. These results seemed to be in agreement with previous conclusions (Section 1.2.3, Chapter 1) that Pd^0-catalyzed allylic isomerisations of 3-deoxy-1-enes resulted in the formation of equilibrium mixtures[38] and as further attempts to change the composition in favour of the desired 19,2-lactone **22** proved to be pointless, the mixture was separated by flash chromatography in order for the allylic bromination to be explored. Surprisingly, treatment of **22** with N-bromosuccinimide in CCl_4 at reflux yielded a complex mixture of compounds and only extensive chromatography afforded a derivative, the ^1H NMR spectrum of which corresponded to structure **37** (6% yield).

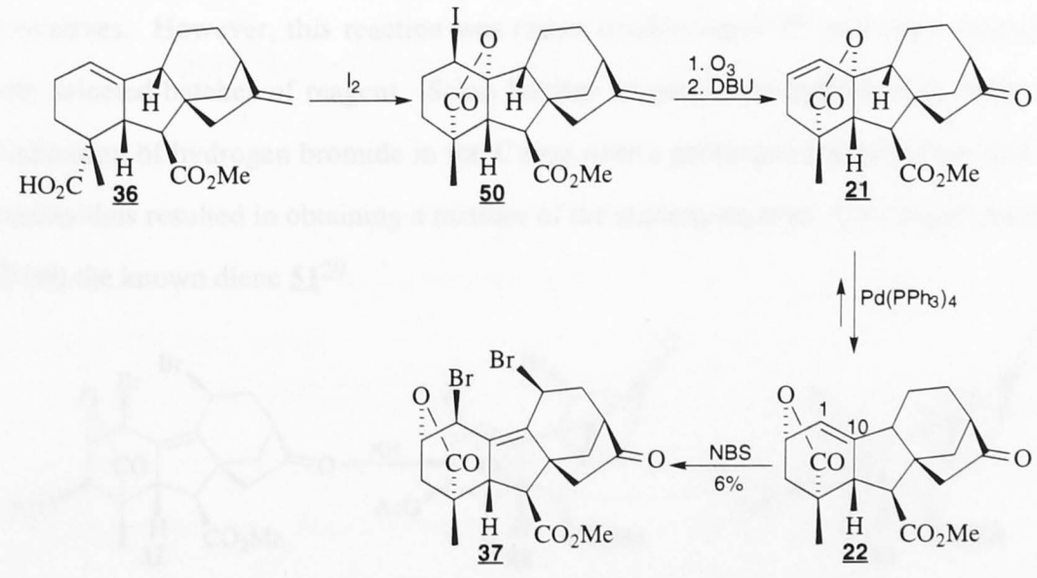


Figure 29. Allylic bromination of 1,(10)-ene 22

The low field region of the spectrum displayed three well-separated signals at 5.11, 4.91 and 4.80 ppm, accounting for one proton each. A doublet of doublets at 4.80 ppm and a doublet at 4.91 ppm were mutually coupled (J = 3.8 Hz) and hence were assigned as H(2β) and H(1α) respectively. A third doublet at 5.11 ppm was assigned as H(11α). The remaining diagnostic resonances of this product (H6, H14α, H14β, H15α, H15β) were in excellent agreement with those of its 3β-AcO congener **17**.

Given this discouraging result, it was clear that the deoxygenation of the 3-hydroxy group could not be carried out in the first part of the sequence due to the apparent stabilizing role of the 3β-acetoxy group in the course of the allylic bromination (**22** *vs* **15**). Further effort was therefore directed towards the partial removal of functionality from the A-ring at a later stage, following the synthesis of Furber[27,29].

2.3.4 Synthesis of 11α- and 11β-hydroxy-9,15-cyclo-GA$_9$ methyl ester

The key intermediate **17** was then synthesized from the GA$_7$ methyl ester (**28**) in 4 steps according to the protocol of Furber[29] and further elaborated towards the desired targets, as outlined in Figure 30. Compound **17** was treated with potassium hydride to effect the intramolecular alkylation, giving access to the 9,15-cyclo derivatives. However, this reaction was rather troublesome[29,60] and only successful with selected batches of reagent. Some batches of potassium hydride also promoted elimination of hydrogen bromide in the C-ring over a prolonged reaction time and the process thus resulted in obtaining a mixture of the starting material, the desired product **29** and the known diene **51**[29].

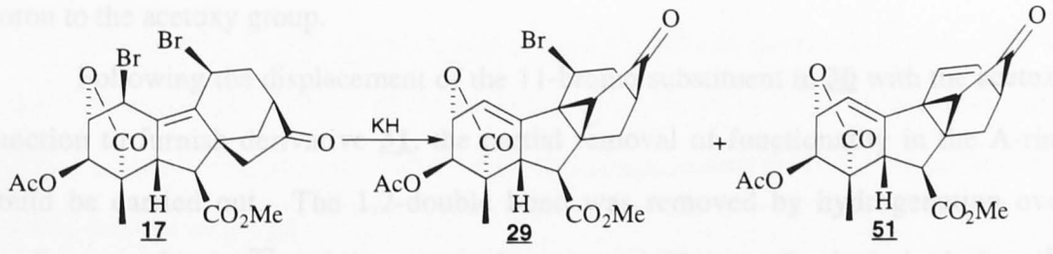


Figure 30. Intramolecular alkylation of 17 with KH

Separation of these three compounds by analytical TLC was not possible and the only way of monitoring the reaction progress was running NMR spectra of the reaction mixture at 6-hour intervals. The treatment of potassium hydride with iodine prior to the reaction did not bring about any significant improvement[60].

However, this problem could be completely circumvented by treating dibromide **17** with dry powdered cesium acetate and 18-crown-6 in benzene at reflux followed by treatment of the product **52** with Me_2BBr (Figure 31). Preliminary experiments of Furber[27,61] had shown that the acetate anion brought about the nucleophilic substitution of bromine at C(11) and was basic enough to enolize the 16-ketone function and initiate alkylation, with the reaction being complete within 45 minutes. In the present work it was found that when the Me_2BBr-mediated rearrangement[29] was attempted on the 19,2-lactone compound **52**, the reaction afforded a single product, the ^1H NMR spectrum of which indicated a loss of one acetoxy group. Further examination of ^1H and ^{13}C NMR spectra revealed that the compound was identical with the 11β-bromo

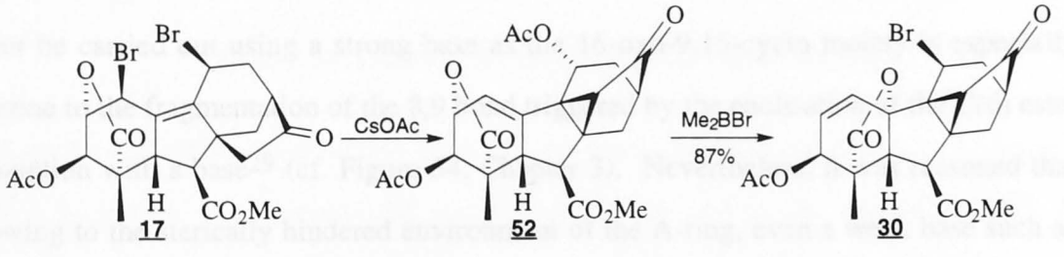


Figure 31. Modified synthesis of the bromo compound 30

derivative **30**. The reagent thus not only reversed the A-ring lactone function, but also displaced the 11-acetoxy group with bromine. This process was obviously facilitated by the neighbouring cyclopropane ring and by the coordination of the highly oxyphilic boron to the acetoxy group.

Following the displacement of the 11-bromo substituent in **30** with the acetoxy function to furnish derivative **31**, the partial removal of functionality in the A-ring could be carried out. The 1,2-double bond was removed by hydrogenation over rhodium on alumina[27] and the saturated compound **32** was selectively hydrolysed[54] with a mixture of potassium carbonate and hydrogen carbonate to the 3-hydroxy deriva-

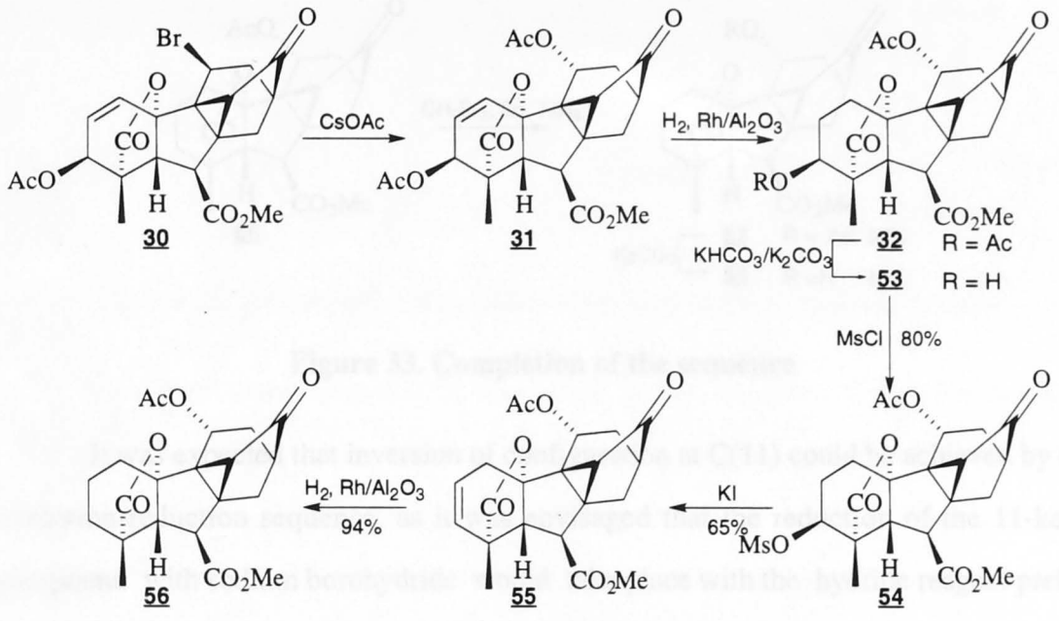


Figure 32. Partial removal of functionality from the A-ring

tive which, without purification, was treated with mesyl chloride to yield the 3-mesylate **54**. The intention was to eliminate MsOH from the A-ring and hydrogenate the resultant 2,3-double bond. It had to be borne in mind, however, that the reaction could not be carried out using a strong base as the 16-oxo-9,15-cyclo moiety is especially prone to the fragmentation of the 8,9 bond triggered by the enolisation of the C(6) ester function with a base[29] (cf. Figure 54, Chapter 3). Nevertheless, it was reasoned that owing to the sterically hindered environment of the A-ring, even a weak base such as iodide would effect an elimination process rather than substitution[62]. This expectation proved to be correct, as reaction with potassium iodide and 18-crown-6 ether in dry DMF furnished the unsaturated product **55**, the 2-ene function of which was hydrogenated over Rh/Al_2O_3 to afford the saturated derivative **56**. As compared to the methods of removing the 1-ene-3-ol moiety in the A-ring which were briefly described in the Introduction to this Chapter, this procedure affords good yields and does not require extensive purification of the material. Restoration of the 17-methylene group by the Lombardo-Oshima procedure[63] and subsequent hydrolysis of the 11-acetate gave the first desired target **58**.

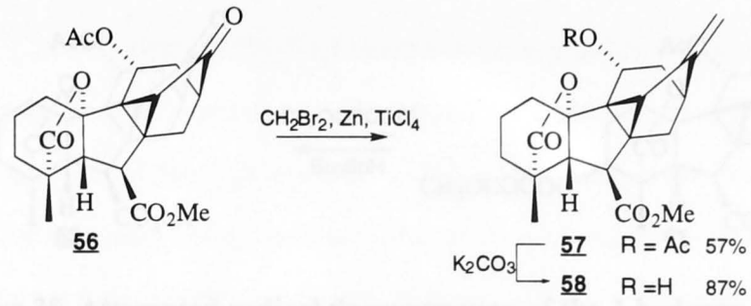


Figure 33. Completion of the sequence

It was expected that inversion of configuration at C(11) could be achieved by an oxidation-reduction sequence, as it was envisaged that the reduction of the 11-keto compound with sodium borohydride would take place with the hydride reagent pref-

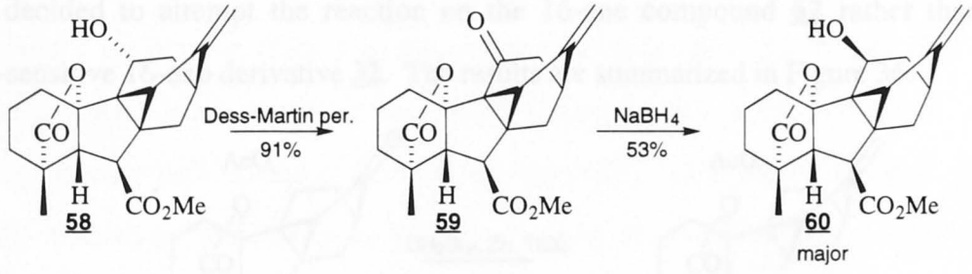


Figure 34. Oxidation/reduction cycle on alcohol 58

erentially attacking the molecule from the less sterically hindered α-face to give rise to the β-epimer. The alcohol **58** was therefore oxidised to the ketone **59** with the Dess-Martin periodinane[64] and subjected to treatment with sodium borohydride. In the event, the reaction gave a mixture of epimers in the ratio of 3:1, with the 11β-carbinol as the major one. Thus, compounds **58** and **60** were obtained from GA_7 methyl ester **28** in 14 and 16 steps respectively.

In order to find a more efficient way of radical deoxygenation of the 3-hydroxy group than that described in the Introduction to this Chapter, alcohol **53** obtained by selective hydrolysis of the diacetate **32** was esterified with methyloxalyl chloride[65]. However, the 3-methyloxalyl derivative **61** did not undergo deoxygenation with Bu_3SnH. Instead, the parent alcohol was reformed, presumably by the addition of Bu_3SnH across the ester carbonyl group followed by decomposition upon work-up[65] (Figure 35).

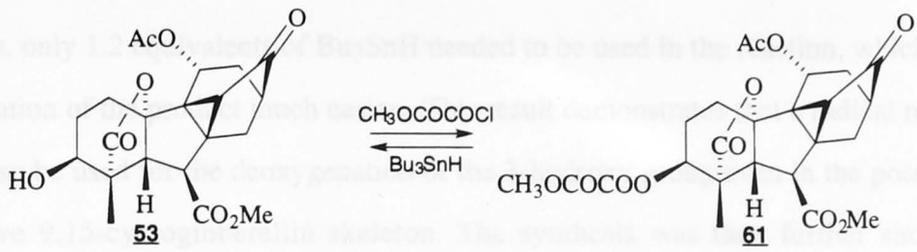


Figure 35. Attempted radical deoxygenation of the 3-hydroxy group

The pentafluorophenoxythiocarbonyl group introduced by Barton and Jaszberenyi[66] appeared to be more suitable, due to the ease with which these esters undergo β-scission. Given the basic conditions needed for successful reaction of a 3β-hydroxy gibberellin derivative with pentafluorophenoxychlorothionoformate[67], it was decided to attempt the reaction on the 16-ene compound **62** rather than the base-sensitive 16-oxo derivative **32**. The results are summarized in Figure 36.

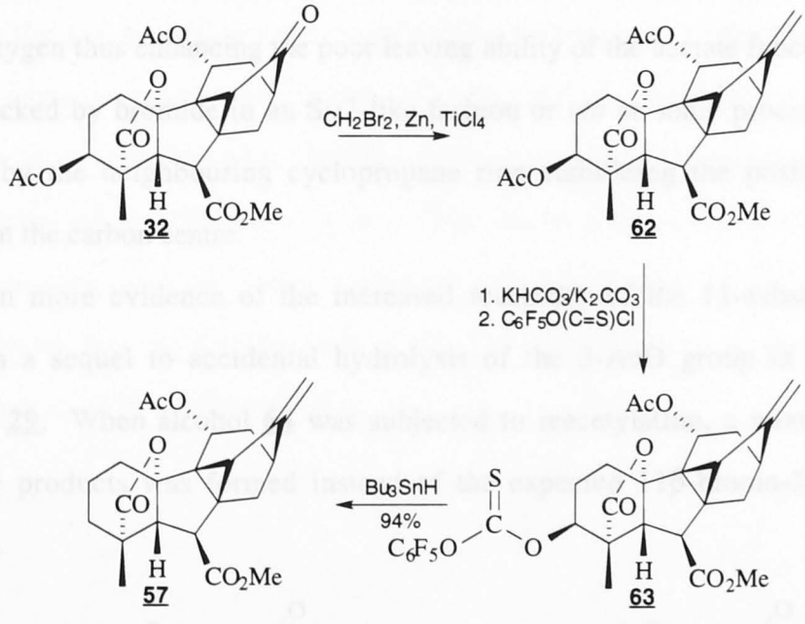


Figure 36. Successfull radical deoxygenation of the 3-hydroxy group

Ketone **32** was converted into the 16-ene **62**[27], which was sequentially treated with a potassium carbonate/hydrogencarbonate mixture[54] to hydrolyse the 3-acetoxy function and then pentafluorophenoxychlorothionoformate to give the Barton-type ester **63**, which underwent smooth radical deoxygenation with Bu_3SnH to furnish **57** in an excellent yield. Compound **57** prepared in this way was identical with an authentic sample synthesized as outlined in Figure 33. Owing to the overall efficiency of the

process, only 1.2 equivalents of Bu_3SnH needed to be used in the reaction, which made purification of the product much easier. This result demonstrates that a radical reaction may also be used for the deoxygenation of the 3-hydroxy group even in the potentially sensitive 9,15-cyclogibberellin skeleton. The synthesis was thus further shortened allowing the target molecules to be prepared in gram quantities, if required.

2.3.5 Reactivity of the 11-substituent in the 9,15-cyclogibberellin skeleton

As described in the previous Section, dimethylboron bromide effected nucleophilic substitution of the 11α-acetoxy group with bromide (**52** → **30**). Although the reaction mechanism was not studied, it can be assumed that boron coordinates to the carbonyl oxygen thus enhancing the poor leaving ability of the acetate function. C(11) is then attacked by bromide in an S_N2-like fashion or *via* an ionic process which is facilitated by the neighbouring cyclopropane ring stabilizing the positive charge developed at the carbon centre.

Even more evidence of the increased reactivity of the 11-substituent was obtained in a sequel to accidental hydrolysis of the 3-AcO group in the bromo compound **29**. When alcohol **64** was subjected to reacetylation, a mixture of two inseparable products was formed instead of the expected 11β-bromo-3-acetate **29** (Figure 37).

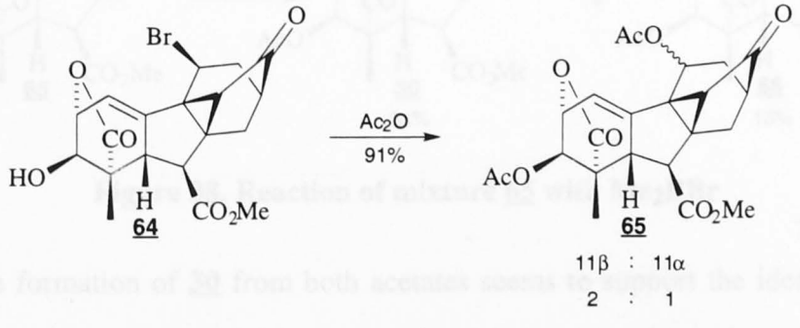


Figure 37. Acetylation of alcohol 64

The ^{1}H NMR spectrum revealed that the signal from H(11α) at 6.23 ppm characteristic of **29** had disappeared and that both of the new compounds bore two acetoxy functions. ^{13}C NMR experiments indicated the presence of two, probably closely related compounds. The mass spectrum displayed an ion at *m/z* 444, the composition of which was found to be $C_{23}H_{24}O_9$, with the next prominent peak being at *m/z* 402 (loss of CH_2CO). This information led to the suspicion that the mixture contained epimeric 11α- and 11β-acetates, formed by the substitution of the 11-bromo function with an acetoxy group. The ^{13}C-^{1}H correlation (HETCOR) spectrum and the comparison of spectroscopic data with those for the previously made 11α-acetate **52** supported this conclusion. The loss of stereochemical integrity of the product coupled with obtaining the 11β-acetate (cf. Figure 38) as the major product suggested that, under these conditions, the reaction took an S_N1 rather than an S_N2-like pathway. The 2:1 ratio of the 11β-acetate to the 11α-acetoxy derivative as determined by NMR did not change when the mixture was resubjected to the reaction conditions for 7 days.

In order to reintroduce the bromine substituent to C(11), the mixture was treated with an excess of Me_2BBr for 4 hours. The reaction afforded stereochemically homogeneous compound **30**, together with some 11β-acetoxy derivative **66** (Figure 38), which indicates that the displacement of the 11α-acetoxy group is faster than the substitution of the 11β-acetoxy function.

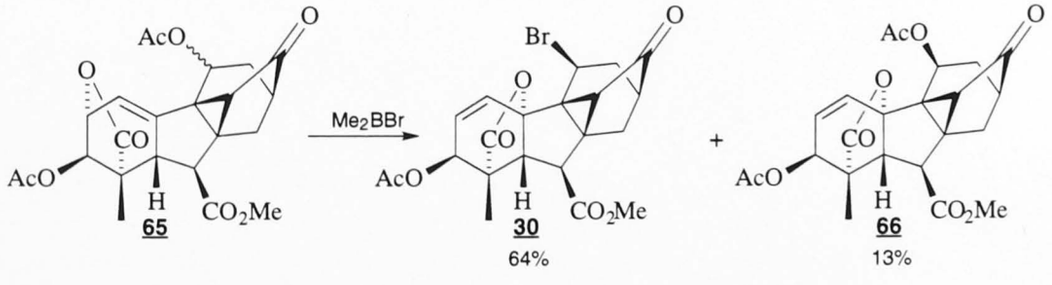


Figure 38. Reaction of mixture 65 with Me_2BBr

The formation of **30** from both acetates seems to support the idea of an S_N1 reaction mechanism. Since further exploration of this area would not be consistent with the primary aim of this thesis, no other experiments on the reactivity of 11-substituted 9,15-cyclo compounds were carried out.

2.3.6 GC-MS analysis

Neither **58** or **60** were identified by GC-MS comparisons as natural products or metabolites. Nevertheless, given the occurrence of 11-hydroxy gibberellins in higher plants, it can be expected that these two synthetic monohydroxy derivatives with the 9,15-cyclogibberellin framework may be isolated during future investigations.

CHAPTER 3

3. SYNTHESES OF 12-HYDROXY-9,15-CYCLOGIBBERELLINS

3.1 INTRODUCTION

Having found that none of the compounds whose structures were to be confirmed by synthesis corresponded to either **58** or **60**, the initial suspicion that the hydroxy group is located at C(12) rather than at C(11) grew even stronger. Attention was thus focused on the development of an efficient synthesis of 12-hydroxy-9,15-cyclogibberellins which would provide access to as many derivatives as possible. The development of such a sequence is the aim of work described in this Chapter.

12-hydroxy gibberellins appear to be among the least accessible derivatives, both in terms of isolation and synthesis[20]. So far, the only synthetic procedure for introducing the hydroxy group to C(12) in GA_9-like gibberellins has been a transannular process utilizing the exocyclic double bond in the D-ring[43,44,45] (Figure 39). The 16-ene function of a given gibberellin derivative is converted into the 16-bromo-17-hydroxy moiety and the 17-hydroxy compound subjected to transannular oxidation with $Pb(OAc)_4$ or $PhI(OAc)_2$. The β-bromoether arrangement of the resultant 16-bromo-12β,17-ether derivative is subsequently cleaved by reductive elimination with zinc in an acidic medium. The 16,17-double bond is thus restored and a 12β-substituent introduced.

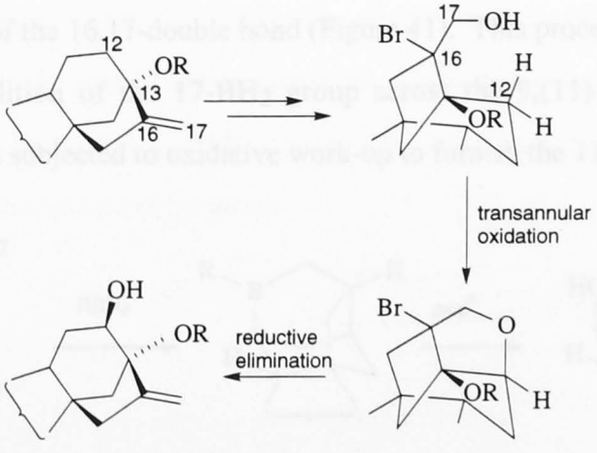


Figure 39. Introduction of the 12-hydroxy function

The presence of the protected 13-hydroxy group is necessary to increase the stability of the intermediate bromohydrin. When this function is replaced with hydrogen, the bromohydrin tends to be unstable and is obtained in a poor yield[44].

Conceptually, this approach does not exclude the possibility of the 9,15-cyclopropyl ring closure. However, it can be estimated that, given the additional steps needed to introduce the cyclopropyl ring, the length of the sequence would be between 25 and 30 steps. Also, using the standard methodology for the introduction of the cyclopropyl ring based on a 1,10-diene acid as the key intermediate would only provide efficient access to the 3-deoxy derivatives, as mentioned in Chapter I. Regardless of these evident drawbacks, this approach was likely to be successful because no apparent difficulties could be foreseen. A synthetic route based on these considerations is described in Section 3.2 of this Chapter.

The above analysis suggested the need for developing another synthetic pathway which would be more flexible and/or shorter. In this regard, the synthesis of 11-hydroxy gibberellins[46] appeared to provide an attractive precedent (Figure 40).

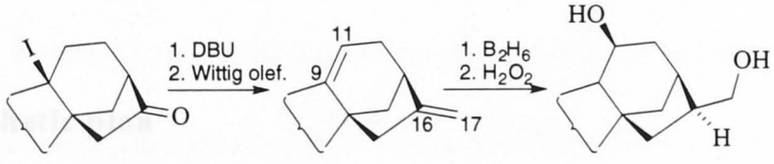


Figure 40. Synthesis of 11-hydroxy gibberellins

Upon hydroboration of a 9(11),16-diene, the borane reagent first adds to the less hindered exo-face of the 16,17-double bond (Figure 41). This process is followed by an intramolecular addition of the 17-BH_2 group across the 9,(11)-ene function. The intermediate is then subjected to oxidative work-up to furnish the 11β,17-diol.

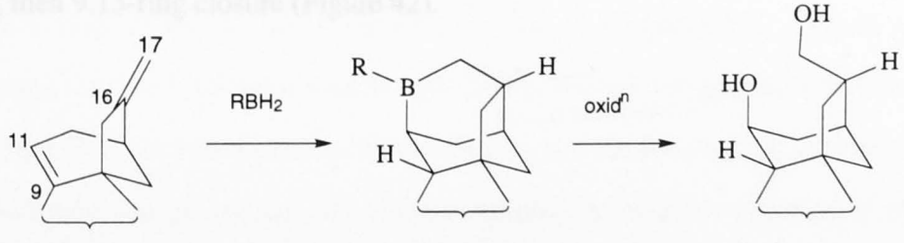


Figure 41. The stereochemistry of hydroboration of 9(11),16-dienes

It is important to note that the exocyclic double bond serves as a stereochemical control element. Although the reaction of borane with the unsymmetrically substituted 9,11-double bond itself could be expected to occur with a reasonable degree of selectivity[68], the initial addition of the reagent to the 16,17-double bond ensures the desired stereocontrol, as the 17-BH_2 group is delivered to the endocyclic double bond on the β-face of the C-ring.

Since the starting 9,(11)-ene compound (a GA_{73} arrangement, see Chapter 1 and Chapter 4) is prepared by the elimination of a 9β-iodo substituent[36,46] (Figure 40), introduced by extending functionality from the A-ring, the process is formally a transposition of synthetic function from C(9) to C(11). The development and completion of a related synthesis of 12-hydroxy-9,15-cycloderivatives from an 11,16-diene system is described in Section 3.3.

3.2 SYNTHETIC ROUTE TO 12-HYDROXY 9,15-CYCLO DERIVATIVES BASED ON TRANSANNULAR OXIDATION

3.2.1 Synthetic plan

In principle, the cyclopropyl ring may be closed before or after the introduction of the OH group to C(12). However, because of the potential sensitivity of the three-membered ring towards the conditions of the transannular process as well as towards the preceding and subsequent reactions, the latter timing seemed to be more secure. The synthesis can therefore be divided into two parts: functionalization of the C-ring, then 9,15-ring closure (Figure 42).

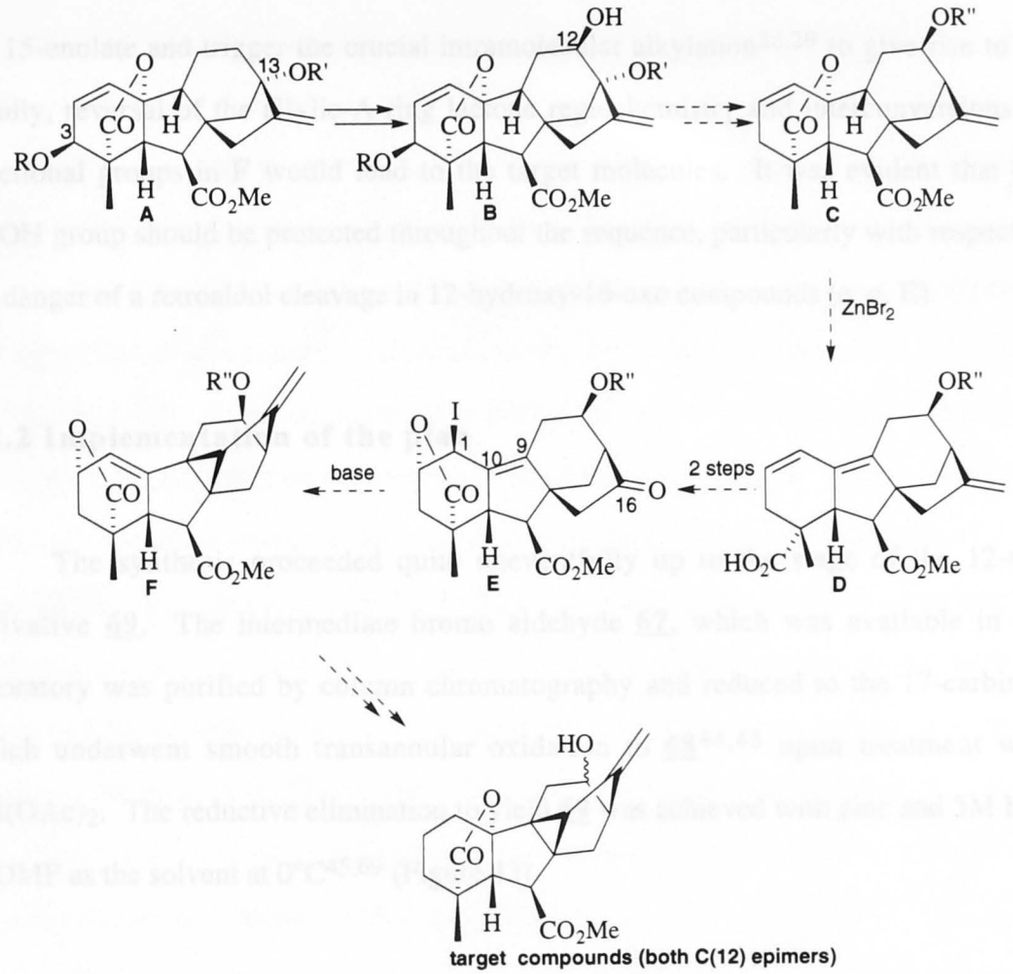


Figure 42. Synthetic plan based on transannular oxidation

The suitably protected GA_3 derivative **A** was the obvious starting material with regard to the role of the 13-substituent in the process of functionalizing C(12) (*vide supra*). The introduction of the 12β-OH group was to be achieved by the transannular oxidation process utilizing the 16,17-double bond in the first part of the synthesis[43,44,45]. Intermediate **B** with the additional OH group was expected to undergo the transformations needed to close the cyclopropyl ring. Deoxygenation of C(13)[65] followed by Li/NH_3 reduction[48] to remove the 3-OR group and subsequent reconstruction of the A-ring[36] would give the 19,10-lactone **C**, which would be rearranged into the 1,10-diene acid **D** with $ZnBr_2$. Given the reported sensitivity of this conversion[36,37] to functionality elsewhere in the molecule, the influence of the 12-substituent was problematical. Upon obtaining **D**, the key compound **E** containing the required 1-halo-16-oxo-9-ene arrangement would be prepared in an iodolactonization-ozonolysis sequence[16,29,36]. Treatment of **E** with base would form

the 15-enolate and trigger the crucial intramolecular alkylation[28,29] to give rise to **F**. Finally, reversal of the allylic A-ring lactone regiochemistry and interconversions of functional groups in **F** would lead to the target molecules. It was evident that the 12-OH group should be protected throughout the sequence, particularly with respect to the danger of a retroaldol cleavage in 12-hydroxy-16-oxo compounds (e. g. **E**).

3.2.2 Implementation of the plan

The synthesis proceeded quite uneventfully up to the stage of the 12-OH derivative **69**. The intermediate bromo aldehyde **67**, which was available in the laboratory was purified by column chromatography and reduced to the 17-carbinol, which underwent smooth transannular oxidation to **68**[44,45] upon treatment with $PhI(OAc)_2$. The reductive elimination to yield **69** was achieved with zinc and 5M HCl in DMF as the solvent at 0°C[45,69] (Figure 43).

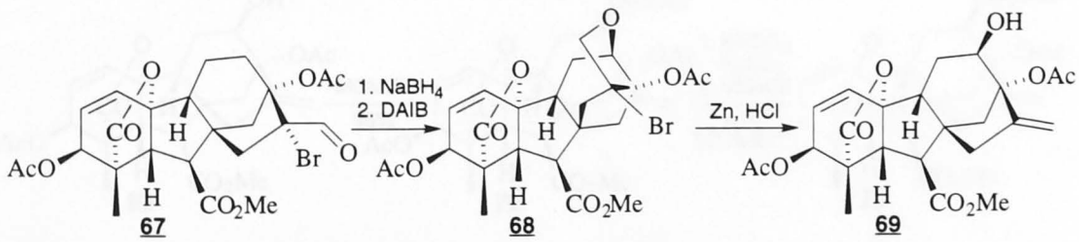


Figure 43. Preparation of the 12-hydroxy derivative 76

The methoxymethyl group was chosen for the protection of the new hydroxy function, due to its excellent stability under various conditions[53b], although its compatibility with the wide array of conditions to be used in the following steps had yet to be established. Not many protective groups can be successfully employed in gibberellin chemistry owing to the sensitivity of these compounds to conditions which may be required during subsequent removal, but the manipulation of the MOM group had been well explored in this laboratory previously[70]. It was expected though, that the protective group would have to be changed at some stage considering that processes in both basic and acidic media were to be carried out. Compound **69** was thus treated with

MOMCl under standard conditions to yield the protected derivative **70**. Selective hydrolysis[54] of the 3-acetate group in **70** was achieved with $KHCO_3/K_2CO_3$ and the 3β-OH function was protected as the MOM ether as a prelude to the Li/NH_3 reduction[36,48] which was to be performed later. In principle, if the 3-acetate group in **69** was deprotected first and the resultant 3,12-di-OH compound treated with MOMCl, the two MOM groups could be introduced in one step. However, there was a possibility that the free 12β-OH group might assist the hydrolysis of the 13-acetoxy function, so these steps were performed sequentially. The 13-acetate group in **71** was hydrolyzed with K_2CO_3 and the crude alcohol esterified with methyloxalyl chloride. Methyloxalyl ester **72** was then deoxygenated[65] with Bu_3SnH to afford the 13-deoxy compound **73**. The structure of compound **73** was corroborated by the disappearance of the methoxyl group (-OCOCOO**Me**) in the ^1H NMR spectrum. The IR spectrum did not show an OH band and the polarity of **73** was lower than that of the parent methyloxalyl compound **72**. The mass spectrum displayed a molecular peak at *m/z* 448.2, its composition ($C_{24}H_{32}O_8$) being consistent with deoxygenation.

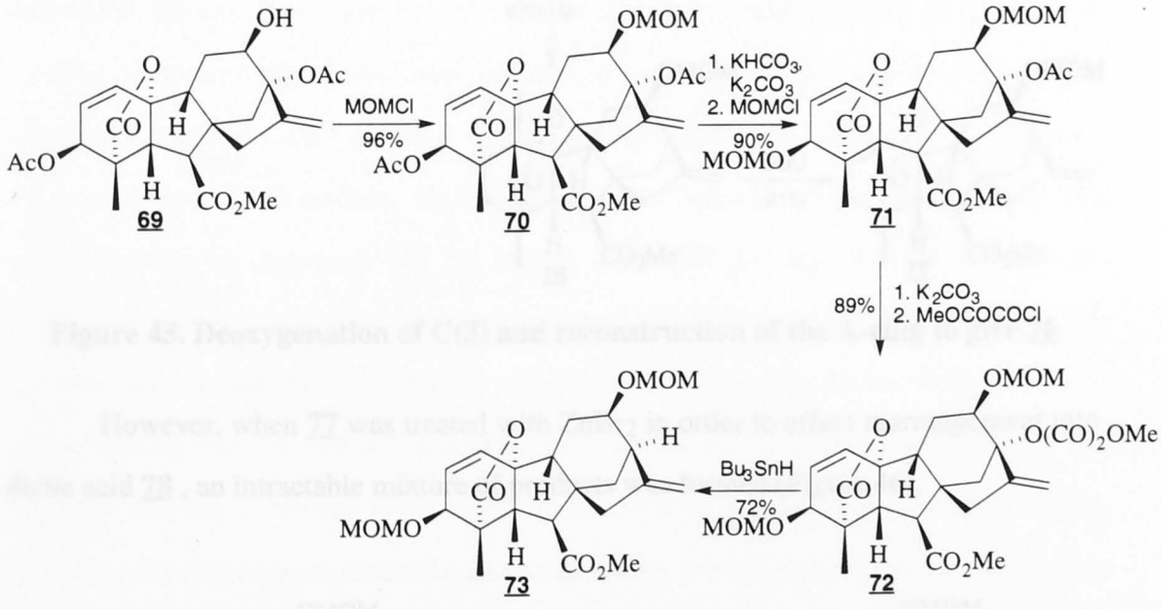


Figure 44. Deoxygenation of C(13)

The synthesis thus reached its second part, where the processes leading to the introduction of the cyclopropyl ring could be implemented. With compound **73** in hand, Li/NH_3 reduction[36,48] was to be attempted first. It could have been argued at this

point that the 13-substituent could also be removed at this stage in a one-pot reaction using the methodology described in Section 2.3.2 of Chapter 2, except that the finding that the 13-mesylate group may be cleaved under the conditions of Li/NH_3 reduction was made after this work had been completed. When **73** was subjected to the reaction conditions, acid **74** was obtained in good yield, together with a small amount of alcohol **75** arising from over-reduction. NMR spectra and mass spectrometry confirmed that the 12-methoxymethoxy group and the C/D ring functionality remained unchanged. The A-ring lactone and 1-ene function were reconstructed in an iodolactonization/ elimination sequence to afford the 3-deoxy compound **77**, as outlined in Figure 45.

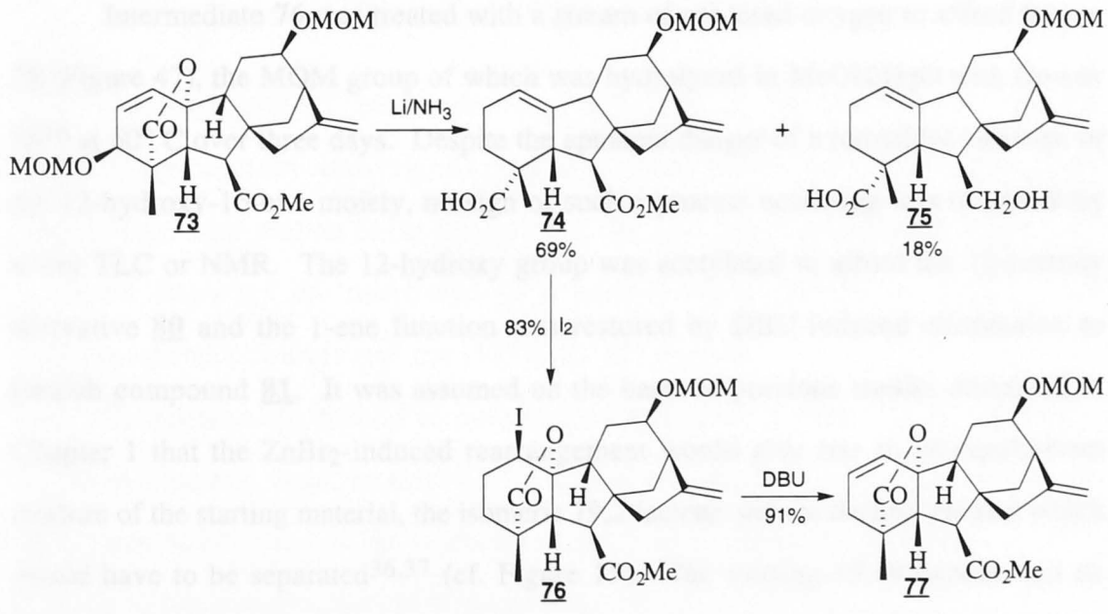


Figure 45. Deoxygenation of C(3) and reconstruction of the A-ring to give 76

However, when **77** was treated with $ZnBr_2$ in order to effect rearrangement into diene acid **78** , an intractable mixture of products was formed (Figure 46).

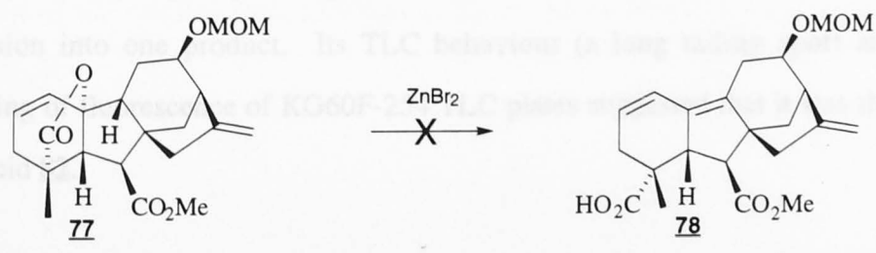


Figure 46. $ZnBr_2$-induced rearrangement attempted on 76

It is known[53b] that MOM and similar protective groups, such as MEM and MTM[53] can be cleaved with Lewis acids, although relatively strong, boron-based acids are needed to cleave a MOM group; nevertheless, the presence of this group was thought to be the reason for the reaction being unsuccessful. It was therefore decided to replace the MOM group with another function. Because of the acidic conditions needed to deprotect the MOM group, it was necessary to remove the acid-sensitive[20] 16,17-double bond first; Dowex 50-catalysed deprotection[70] of **77** attempted on a small scale afforded a complex mixture of products by TLC. The logical consequence of these considerations was the following timing of synthetic steps: ozonolysis (before reintroducing the 1,2-double bond) /deprotection /reprotection.

Intermediate **76** was treated with a stream of ozonised oxygen to afford ketone **79** (Figure 47), the MOM group of which was hydrolyzed in MeOH/H_2O with Dowex 50[70] at 60° C over three days. Despite the apparent danger of a retroaldol cleavage of the 12-hydroxy-16-oxo moiety, no sign of such a process occurring was observed by either TLC or NMR. The 12-hydroxy group was acetylated to afford the 12-acetoxy derivative **80** and the 1-ene function was restored by DBU-induced elimination to furnish compound **81**. It was assumed on the basis of previous studies described in Chapter 1 that the $ZnBr_2$-induced rearrangement would give rise to an equilibrium mixture of the starting material, the isomeric 19,2-lactone and the desired product which would have to be separated[36,37] (cf. Figure 15). The starting 19,10-lactone and its 19,2-counterpart would then be resubjected to the reaction conditions. When the reaction was carried out under strictly anhydrous conditions, the formation of a mixture of the starting material and two products was observed by TLC. However, allowing the reaction mixture to absorb atmospheric moisture slowly ($ZnBr_2$ is very hygroscopic itself) so that the reagent gradually dissolved over a few hours, resulted in a fast clean conversion into one product. Its TLC behaviour (a long tailing spot) and strong quenching of fluorescence of KG60F-254 TLC plates suggested that it was the desired diene acid **82**.

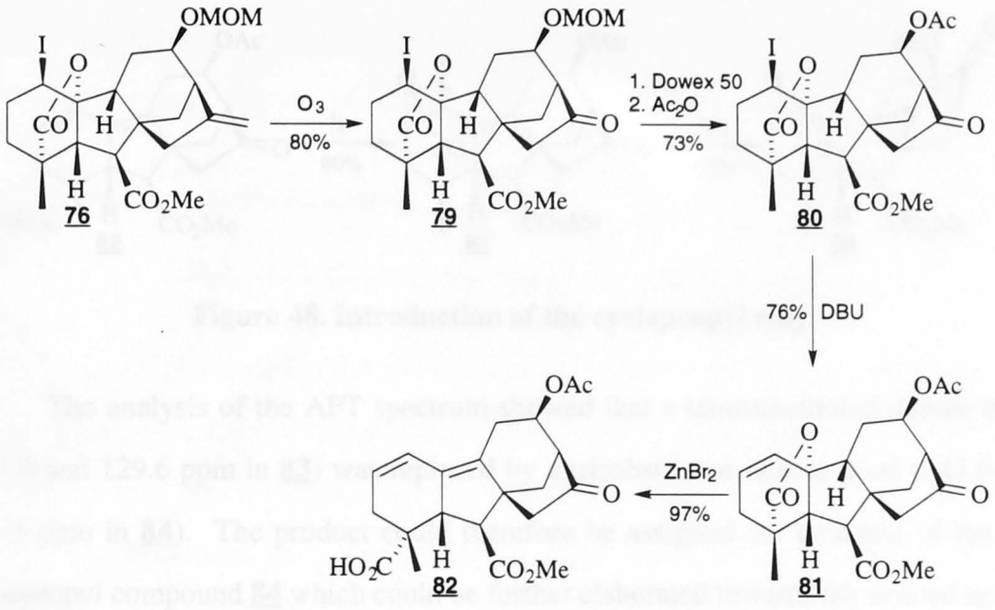


Figure 47. Conversion of 76 into the 1,10-diene acid 82

^{1}H NMR and ^{13}C NMR spectra were fully in accord with structure **82**. The ^{1}H NMR spectrum contained signals assignable to H(1) and H(2) at 5.84 and 6.22 ppm, respectively, their profiles being indicative of the 1,10-diene arrangement[36]. The APT spectrum displayed the resonances of a tetrasubstituted double bond (132.5 and 130.2 ppm) and a disubstituted one (129.9 and 121.5 ppm).

It appears, then, that the formation of the previously reported[36,37,38] equilibrium mixtures obtained upon treatment of 1-ene-19,10-lactone-16-oxo derivatives with $ZnBr_2$ can be attributed to the solubility of the reagent rather than to sensitivity of the reaction to functionality elsewhere in the molecule.

Having obtained the crucial diene acid **82**, the stage could be set for the intramolecular alkylation to close the cyclopropyl ring. **82** was cyclized with iodine to afford iodolactone **83** (Figure 48), which underwent a fast reaction with potassium hydride at 0°C to afford a single product, the ^{1}H NMR spectrum of which revealed that the signal of H(1) shifted from 5.05 ppm in **83** downfield to 6.00 ppm in the product.

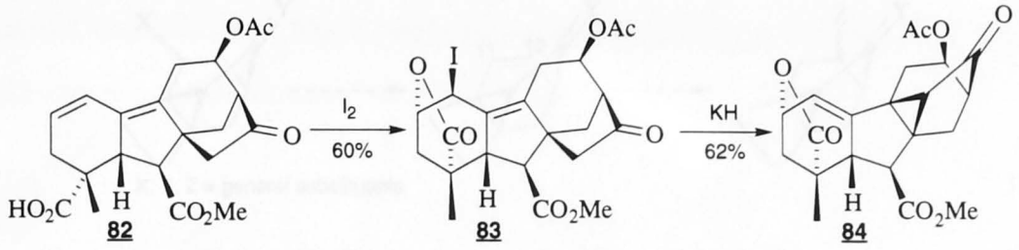


Figure 48. Introduction of the cyclopropyl ring

The analysis of the APT spectrum showed that a tetrasubstituted double bond (143.0 and 129.6 ppm in **83**) was replaced by a trisubstituted double bond (147.0 and 121.0 ppm in **84**). The product could therefore be assigned the structure of the key cyclopropyl compound **84** which could be further elaborated towards the desired targets using functional group interconversion reactions. At this point, the amount of compound **84** available to complete the synthesis was 12 mg and, consequently, more material would have had to be prepared. Since the second approach involving 1,2 functional group transposition had started to bring encouraging results, further work along the present route was suspended.

3.3 SYNTHESIS BASED ON THE FORMAL TRANSPOSITION OF THE 11-SUBSTITUENT TO C(12)

3.3.1 Synthetic plan

The availability of 11-substituted-9,15-cyclogibberellins in gram quantities and the precedent[46] (hydroboration of gibberelline 9(11),16-dienes) described in the Introduction to this Chapter led directly to the conceptual basis for this approach: if the 11-substituent in a suitable 9,15-cyclo compound could be efficiently eliminated, then functionalisation of the resultant 11,12-double bond may produce a C(12)-substituted derivative (Figure 49).

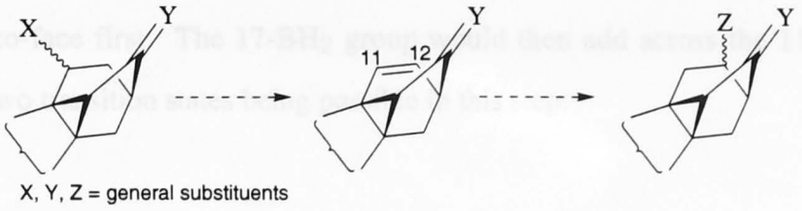


Figure 49. General concept of the synthetic plan

Electrophilic addition did not seem to be a suitable process as it could well be expected that, after the activation of the double bond by an electrophile (e.g. Br^+), the nucleophile (e.g. H_2O) would attack the electron-deficient C(11) (see Section 2.3.5, Chapter 2). Moreover, such a reaction, regardless of its course, would introduce a substituent to C(11) as well and replacement of this function with hydrogen would be necessary at the next stage of the synthesis. Since achieving this would most likely involve a radical reduction, the considerable danger of cyclopropyl ring fragmentation would arise, as shown by Furber and Mander[29] (Figure 50).

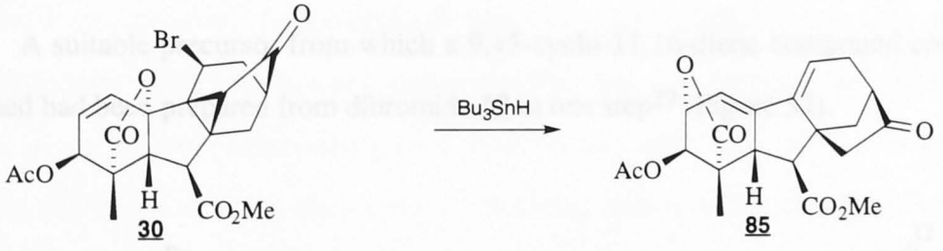
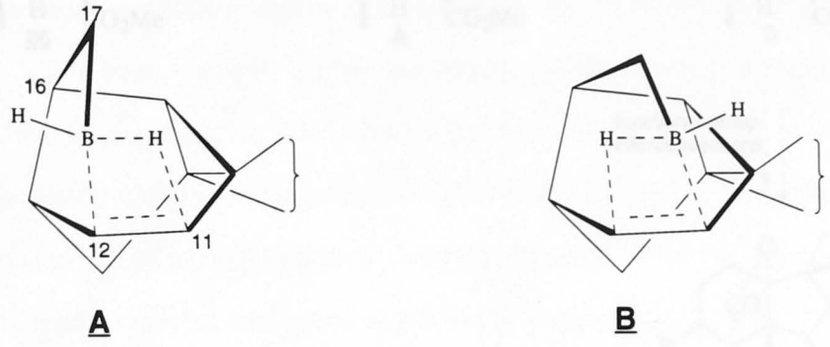


Figure 50. Fragmentation of the cyclopropyl ring *via* a radical centre at C(11)

In contrast, the same polarization of the double bond induced by the cyclopropyl ring makes it quite suitable for the addition of an electron-deficient reagent, such as diborane[68]. Considering the properties of C(11) and C(12), a borane reagent would be likely to attack C(12) with the electron-deficient boron atom and the hydride part of the reagent would become attached to C(11). The intermediate would, after an oxidative work-up, give a compound with a 12-hydroxy group. The selectivity of the overall process might be further increased, if the exocyclic 16,17-double bond was used as a control element. Similar to the example[46] described in the Introduction to this Chapter, the borane reagent would attack the 16-ene function predominantly from the less

hindered exo-face first. The 17-BH_2 group would then add across the 11,12-double bond with two transition states being possible in this step:

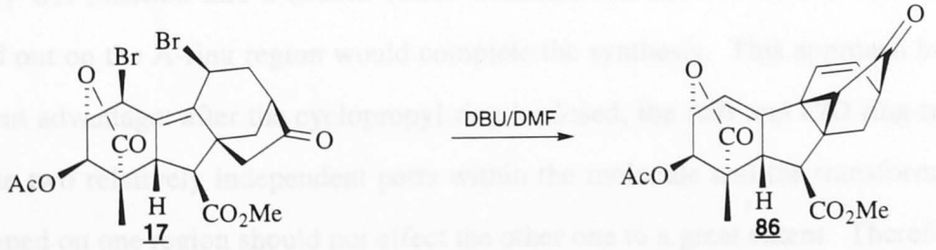


A **B**

Figure 51. Transition states for the intramolecular borane addition

It was presumed that the transition state **A** was likely to be favoured over **B** for two reasons: due to the above-mentioned electronic control, and secondly, the rigid geometry of the C/D ring region imposing greater strain on the intermediate arising through transition state **B**.

A suitable precursor from which a 9,15-cyclo-11,16-diene compound could be obtained had been prepared from dibromide **17** in one step[29] (Figure 52).



17 **86**

Figure 52. A convenient synthesis of an 11-ene intermediate

The synthetic plan thus becomes very straightforward (Figure 53). After the 16-ene function is restored to give **A**, the crucial hydroboration step could be attempted.

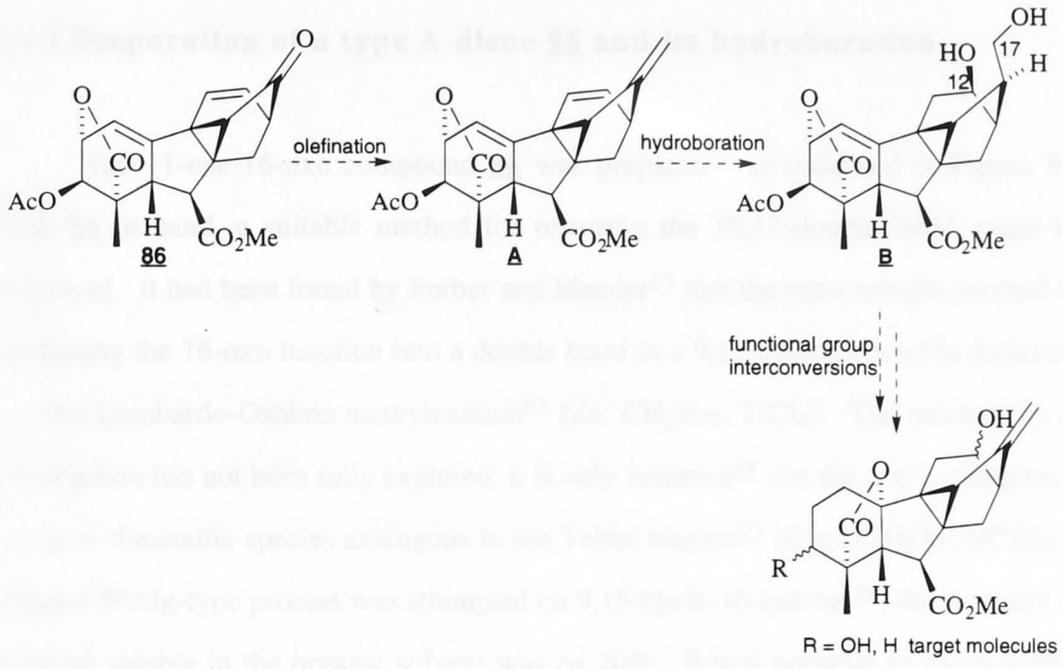


Figure 53. The final plan of the synthesis

The 1,10-double bond is quite inaccessible for steric reasons and it could be expected that, under suitable conditions, its reaction with the borane would not be a significant side process. Intermediate **B**, obtained upon oxidative work-up from the initially formed 12β,17-boracyclo compound, would be elaborated towards the desired targets *via* a protection/deprotection/elimination sequence in order to convert the primary OH function into a double bond. Standard reactions described in Chapter 1 carried out on the A-ring region would complete the synthesis. This approach has one apparent advantage: after the cyclopropyl ring is closed, the A/B and C/D ring regions become two relatively independent parts within the molecule and the transformations performed on one region should not affect the other one to a great extent. Therefore, as compared to the original pathway, where the 3-OH group in the A-ring had been removed in order to allow an efficient synthesis of the cyclopropyl ring, this route would allow the synthesis of the 3-hydroxylated series as well.

3.3.2 Preparation of a type A diene 96 and its hydroboration

The 11-ene-16-oxo compound **86** was prepared[29] as indicated in Figure 52. With **86** in hand, a suitable method for restoring the 16,17-double bond could be employed. It had been found by Furber and Mander[16] that the most reliable method of converting the 16-oxo function into a double bond in a 9,15-cyclogibberellin molecule was the Lombardo-Oshima methylenation[63] (Zn, CH_2Br_2, $TiCl_4$). The mechanism of this reaction has not been fully explored; it is only assumed[63] that the active component is a gem-dimetallic species analogous to the Tebbe reagent[71] ($Cp_2TiCH_2AlCl(CH_3)_2$). When a Wittig-type process was attempted on 9,15-cyclo-16-ketones[16], the recovery of material soluble in the organic solvent was *ca* 20%. It was possible to glean further amounts of the olefin by heating the aqueous layer with DBU, which suggested the formation of a stable, water-soluble salt or betaine. It was also observed[16] that under the basic Wittig conditions the 9,15-cyclo-16-oxo moiety underwent fragmentation already mentioned in Chapter 2 (Figure 54).

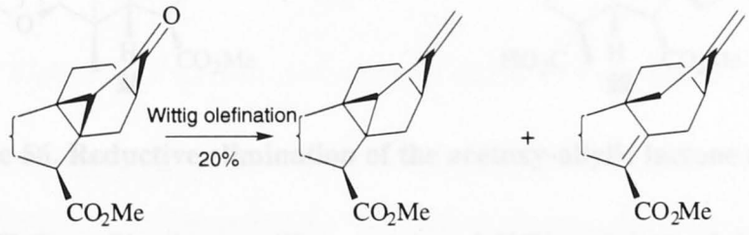


Figure 54. Wittig-type methylenation on a 9,15-cyclo-16-oxo derivative

The yields of the Lombardo-Oshima reaction, when applied to gibberellins, are highly variable[16,24,57,63] (from 40 to 80%). Generally, more functionality on the basic skeleton gives rise to lower yields and *vice versa*. When ketone **86** was subjected to the standard reaction conditions, the reaction failed completely (Figure 55).

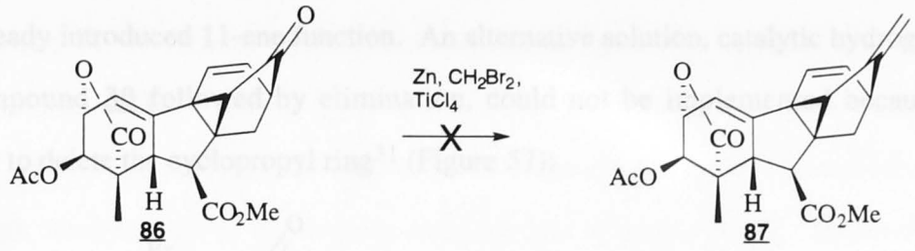


Figure 55. Attempted Lombardo-Oshima methylenation of compound 86

Although the formation of olefinic products was observed by TLC, a number of polar byproducts was also detected, their intensity increasing with time. TLC and NMR monitoring revealed that the reaction afforded a complex mixture of polar compounds, the nature of which was not further analysed. The desired diene product **87** was not detected at all. The appearance of highly polar products can possibly be explained by the observation made by Chu[44] that upon treatment of **88** with Zn in acetic acid 1(10),2-diene acid **89** was the main product (Figure 56).

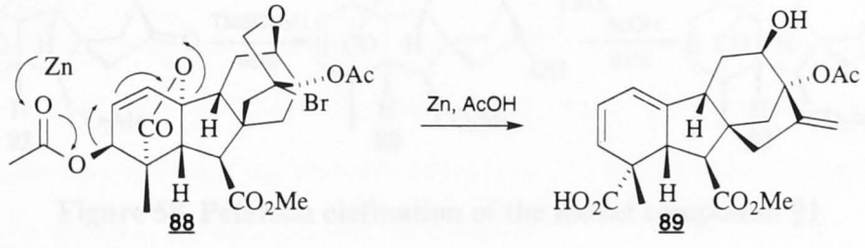


Figure 56. Reductive elimination of the acetoxy-allylic lactone moiety

The TLC profile (long, tailing spots) and "UV activity" of the compounds supported the idea that a similar process took place in this case. These initial products were probably rapidly destroyed by the reagent, as suggested by the ^1H NMR spectrum of the crude reaction mixture. The reaction of **86** with a related system[72] using Cp_2ZrCl_2 instead of $TiCl_4$ turned out to be just as disastrous. The reason for these failures seemed to be the presence of the allylic 1-ene moiety in the A-ring, because the Lombardo-Oshima procedure had been successfully applied[16,24,57,63] to gibberellins with no A-ring allylic arrangement. It appears that if an allylic ene-function in the A-ring is present, the ring becomes sensitive to certain combinations of reagents which may trigger reductive eliminations analogous to that shown in Figure 56. Unfortunately, the 1,10-double bond in **86** could not be removed at this stage because of

the already introduced 11-ene function. An alternative solution, catalytic hydrogenation of compound **30** followed by elimination, could not be implemented because it is known to delete the cyclopropyl ring[31] (Figure 57):

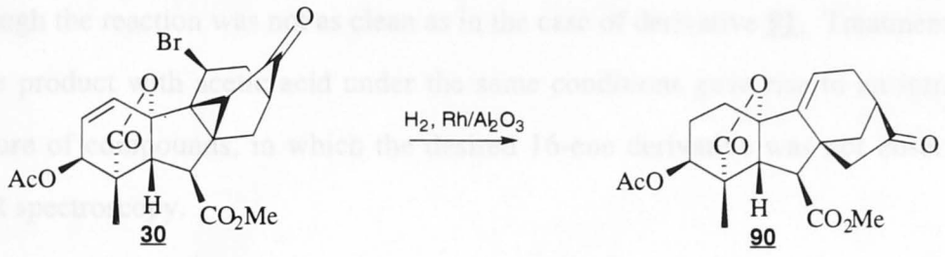


Figure 57. Hydrogenation of compound 30

An attempt was therefore made to carry out the Peterson elimination procedure[73,74]. Due to the potentially base-sensitive methoxycarbonyl and lactone functions, the reaction was performed on a model compound **91** first (Figure 58).

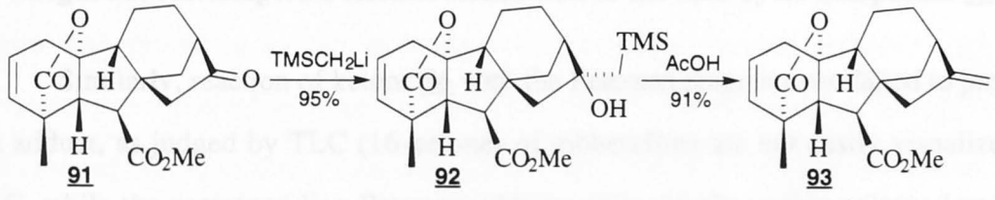


Figure 58. Peterson olefination of the model compound 91

Ketone **91** gave an excellent yield of adduct **92** with $TMSCH_2Li$ at -78°C. The structure of **92** was corroborated by the ^1H NMR spectrum, which displayed an intense singlet of the TMS group at 0.05 ppm, and from the mass spectrum. The stereochemistry of the newly formed stereocentre at C(16) was dictated by the bulky nucleophile attacking the less hindered exo-face of the 16-oxo function. Adduct **92** turned out to be a very stable compound. The decomposition into olefin **93** was effected by heating a solution of **92** in glacial acetic acid at 50°C over several days. The same process in THF with a few drops of concentrated sulphuric acid was much faster by TLC. However, the reaction was not clean and NMR analysis of the crude reaction mixture revealed the presence of the starting material and the expected compound **93**. It also showed a signal at 5.49 ppm, indicating the migration[20] of the 16-ene function into the D-ring. Having established the validity of the Peterson

olefination in the model system, the procedure could be applied to 9,15-cyclo compounds. When ketone **32** , available in good quantities was treated with $TMSCH_2Li$ (Figure 59), the formation of the expected adduct was observed by TLC, although the reaction was not as clean as in the case of derivative **91**. Treatment of the crude product with acetic acid under the same conditions gave rise to an intractable mixture of compounds, in which the desired 16-ene derivative was not observed by NMR spectroscopy.

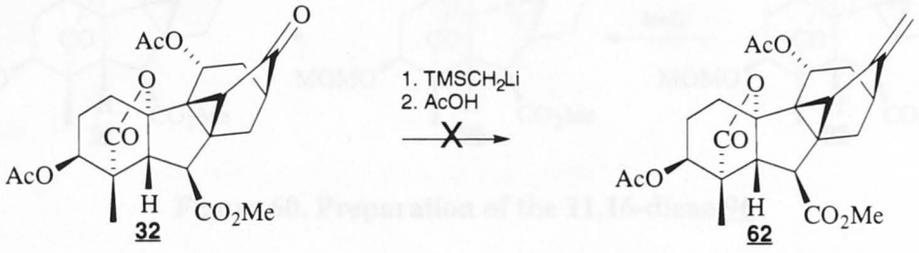


Figure 59. Attempted Peterson olefination of the 9,15-cyclo compound 32

Similarly, reaction of ketone **86** with the Peterson reagent even failed to produce the adduct, as judged by TLC (16-ketones of gibberellins are not easily visualized on TLC, while the corresponding Peterson adducts were clearly visible coloured spots of higher R_f upon spraying with sulphuric acid/vanillin).

These results, however discouraging, did not constitute a serious reason for ceasing to continue further work along this pathway. It was felt that at least the hydroboration of a suitable 11,16-diene derivative should be attempted in order to assess the future prospects of the concept, and attention was logically focused on ketone **32**. Although the preparation of **32** required more steps than the synthesis of **86**, its reaction with the Lombardo-Oshima reagent proceeded successfully[27] and discrimination between the two acetoxy groups could be effected by hydrolysis (see Chapter 2, Section 2.3.4). Another obvious bonus of utilizing compound **32** was that its A/B ring region was consistent with the desired targets. The problem was thus formally reduced to the removal of the 11α-acetoxy function by means of an elimination process (Figure 60).

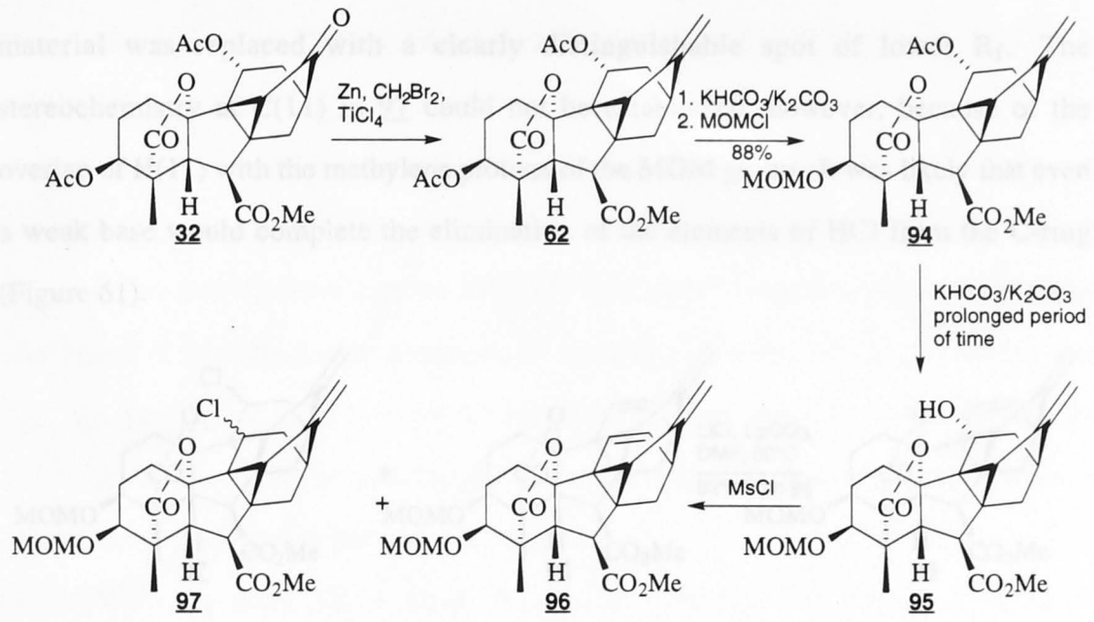


Figure 60. Preparation of the 11,16-diene 96

Diacetate **62** was selectively hydrolysed with $KHCO_3/K_2CO_3$ and the 3-hydroxy group was protected as the methoxymethyl ether to furnish derivative **94**. The 11-acetoxy group in **94** was then saponified with the same combination of reagents over a prolonged period of time. The crude alcohol **95** was treated with mesyl chloride and triethylamine in order to convert the 11-hydroxy function into a leaving group. The reaction seemed to have afforded one major product by TLC, but careful examination of TLC behaviour followed by NMR analysis revealed that the product was a mixture of two compounds. The presence of a triplet at 6.05 ppm and a doublet at 6.10 ppm in the ^1H NMR spectrum indicated that one of the components was the desired diene **96**. The two signals were assigned as H(12) and H(11), respectively and the ^{13}C NMR spectrum, exhibiting resonances at 129.0 ppm and 121.4 ppm, strongly supported this conclusion. It seemed likely that the other compound was the 11-chloro compound **97** formed by nucleophilic substitution of the 11-MsO group by chloride ion (when the mixture was chromatographed on silica gel, a spot of the same R_f as the starting alcohol was detected by TLC in all fractions containing **96** and **97**, although it was not observed in the crude reaction mixture before and after work-up; this suggested that the 11-chloro compound was partially hydrolyzed during chromatography). A control reaction of 0.5 mg of the mixture of **96** and **97** with NaI in acetone indeed showed that part of the

material was replaced with a clearly distinguishable spot of lower R_f. The stereochemistry at C(11) in **97** could not be established, however, because of the overlap of H(11) with the methylene protons of the MOM group. It was likely that even a weak base would complete the elimination of the elements of HCl from the C-ring (Figure 61).

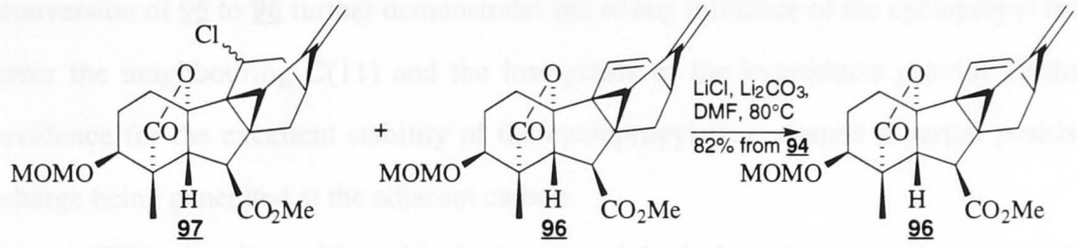


Figure 61. Completion of the synthesis of diene 96

The mixture was dissolved in dry DMF and heated at 80°C with LiCl and Li_2CO_3[75]. As expected, a fast reaction afforded the desired diene **96** in excellent yield. Two minor products were isolated in about 6% yield, arising from attack at C(17) (path a) and C(15) (path b), respectively, with concomitant fragmentation of the cyclopropyl ring (Figure 62).

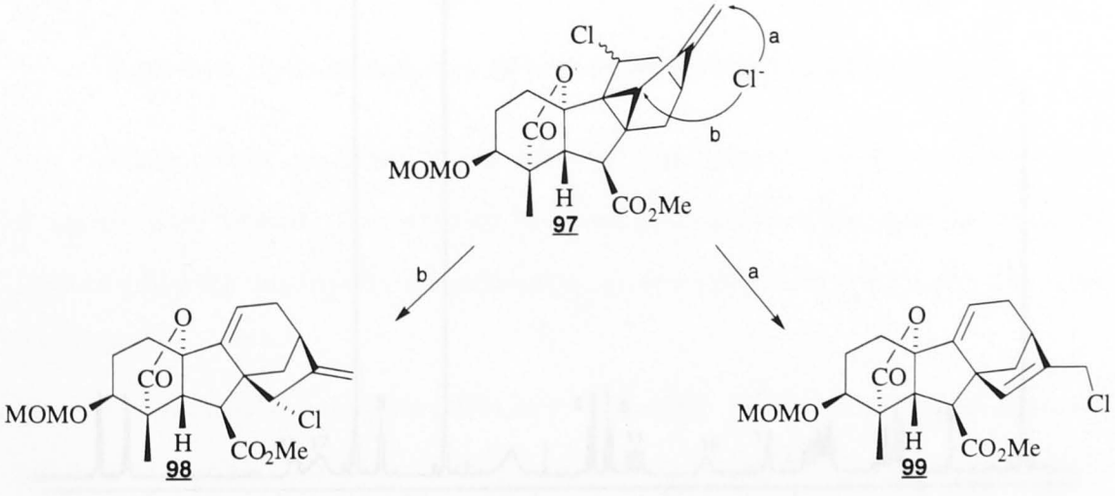


Figure 62. Minor products from the final step of the synthesis of the the diene 96

Compounds **98** and **99** formed an inseparable mixture; the mass spectrum displayed an ion at at *m/z* 422, the composition of which was found to be $C_{22}H_{27}O_6{}^{35}Cl$, the next significant ion being at *m/z* 387 (loss of Cl). ^1H NMR spectrum displayed resonances from **99** at 6.10 ppm (s, H15) and 5.72 ppm (m, H11).

Diagnostic signals of **98** were found at 5.96 ppm (m, H11), 5.27 ppm (s, H17) and 5.32 ppm (s, H'17). The stereochemistry at C(15) in **98** was a consequence of the S_N2 mode of nucleophilic attack (an alternative S_N1 process initiated by dissociation of Cl^- from C(11) followed by fragmentation and finally completed by nucleophilic attack at newly created cationic centres at C(15) and C(17) is also a possibility). The conversion of **95** to **96** further demonstrates the strong influence of the cyclopropyl ring over the neighbouring C(11) and the low yields of the byproducts provide further evidence for the excellent stability of the cyclopropyl ring, despite a partial positive charge being generated at the adjacent carbon.

With the diene **96** in hand, the crucial hydroboration procedure could be attempted. The reaction of **96** with the borane-dimethyl sulfide complex[68] was carried out at 0°C in order to achieve maximum exo/endo selectivity in the initial addition across the 16,17-double bond. After an oxidative work-up, it gave one major, highly polar product accompanied by two byproducts (**A** and **B**, see Section 3.3.6) and some recovery of the starting material. Preliminary ^1H NMR studies on the major compound

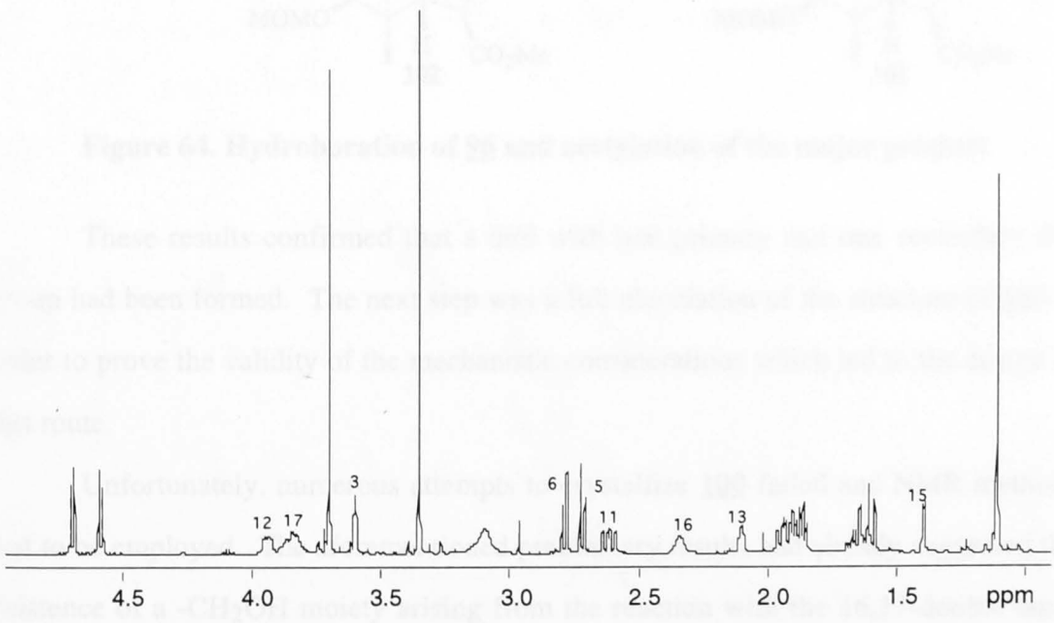


Figure 63. 500 MHz ^1H NMR spectrum of the major hydroboration product (selected resonances are labelled with the position number)

showed the disappearance of both double bonds and the appearance of two new multiplets at 3.95 ppm (one H) and at 3.85 ppm (two H's). Carefully controlled

exposure of this material to acetic anhydride yielded two compounds, the ^1H NMR analysis of which revealed that in the more polar compound the multiplet at 3.85 ppm shifted to 4.42 ppm and became a doublet, while the multiplet at 3.95 ppm remained in its original position; only one acetyl group was observed. The less polar derivative contained two acetyl groups and both multiplets shifted downfield, the two-proton signal becoming a doublet again (Figure 64).

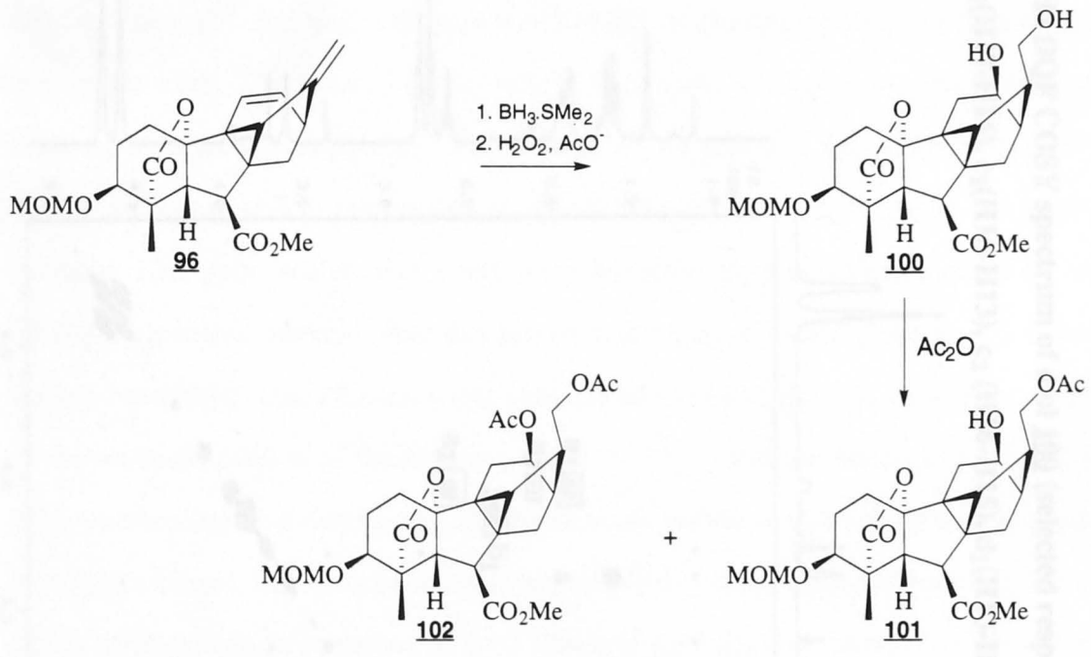
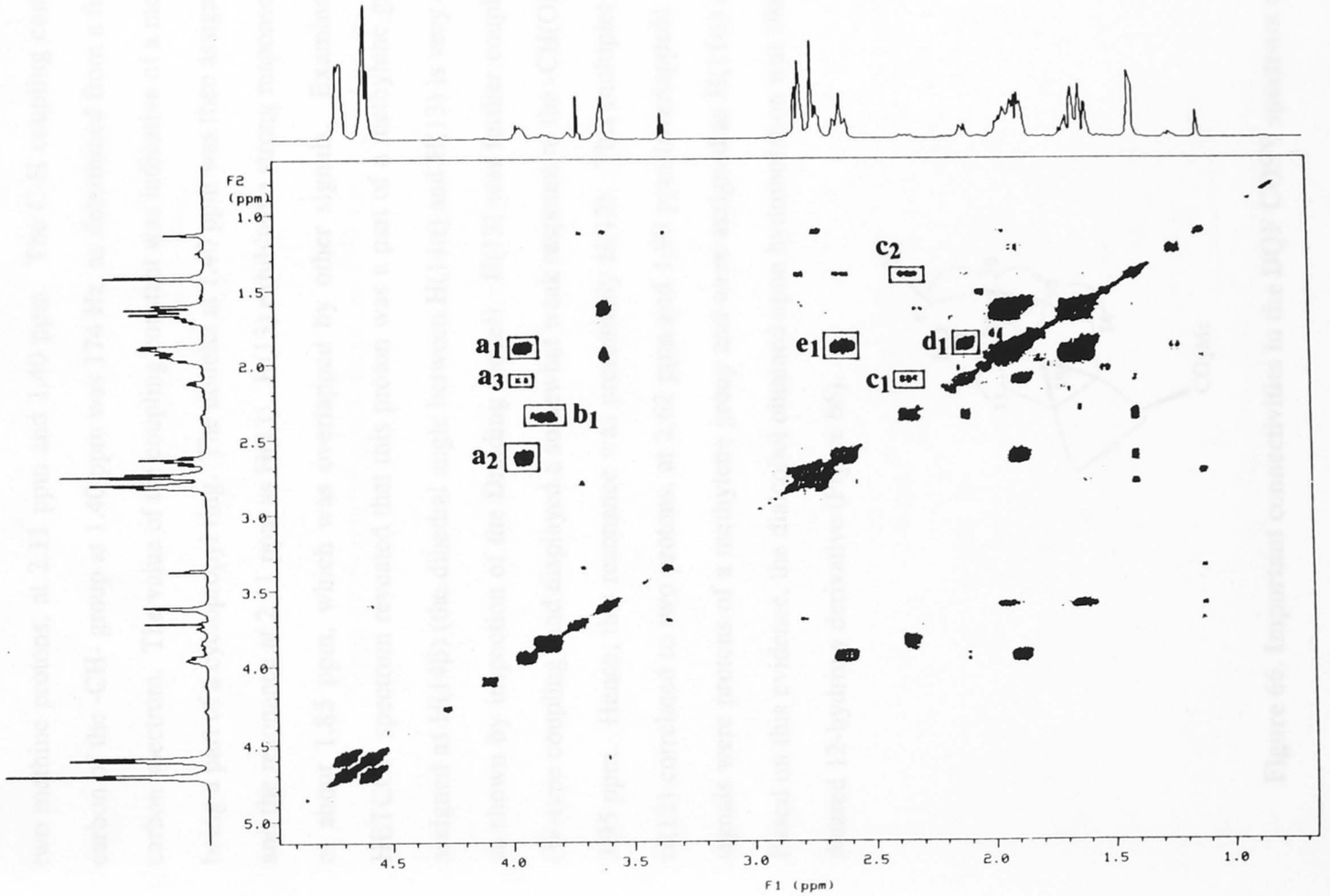


Figure 64. Hydroboration of 96 and acetylation of the major product

These results confirmed that a diol with one primary and one secondary OH group had been formed. The next step was a full elucidation of the structure of **100** in order to prove the validity of the mechanistic considerations which led to the design of this route.

Unfortunately, numerous attempts to crystallize **100** failed and NMR methods had to be employed. The aforementioned preliminary results had already suggested the existence of a $-CH_2OH$ moiety arising from the reaction with the 16,17-double bond and a -CH(OH)- arrangement formed upon further reaction with the 11-ene function. The mass spectrum showed a strong peak at *m/z* 404, the composition of which was found to be $C_{22}H_{28}O_7$, indicating a loss of H_2O from the molecular ion. Although M^+ was detected as well, it was too weak to allow the measurement of its exact mass.

Figure 65. 500 MHz DQF COSY spectrum of diol 100 {selected responses: a_1 (H12α-H11β), a_2(H12α-H11α), a_3(H12α-H13), b_1(H17-H16), c_1(H16-H13), c_2 (H16-H15), d_1(H13-H14β), e_1(H11α-H11β)}.



NMR studies started with a combination of DQF COSY, HETCOR and DEPT experiments[76]. The multiplet at 3.85 ppm which had already been assigned as H(17) and H'(17) was correlated with a multiplet at 2.35 ppm in the DQF COSY spectrum (Figure 65). This signal accounted for one proton and correlated to a methine carbon in the HETCOR spectrum. It was therefore assumed to be H(16). H(16) was coupled to two methine protons, at 2.11 ppm and 1.40 ppm. The C-H coupling constant of the carbon of the -CH- group at 1.40 ppm was 174 Hz, as determined from a fully coupled carbon spectrum. The value of the coupling constant was indicative of a methine group being a part of a cyclopropyl ring. The doublet at 1.40 ppm was then assigned as H(15) and the resonance at 2.11 ppm as H(13). H(13) exhibited a strong response to a proton at about 1.85 ppm, which was overlapped by other signals. Examination of the HETCOR spectrum revealed that this proton was a part of a methylene group; it was assigned as H(14β) (the dihedral angle between H(14α) and H(13) is very close to 90° as shown by inspection of the Dreiding model). H(13) was further coupled to H(15) (W-type coupling) and displayed a somewhat weak response to the -CH(OH)- group at 3.95 ppm. Hence, this resonance was presumably H(12). This multiplet assigned as H(12) correlated to two protons, at 2.62 ppm and 1.90 ppm (overlapped). These two signals were protons of a methylene group and were assigned as H(11α) and H(11β). Based on this evidence, the dialcohol obtained upon hydroboration was likely to be the desired 12-hydroxy derivative (Figure 66).

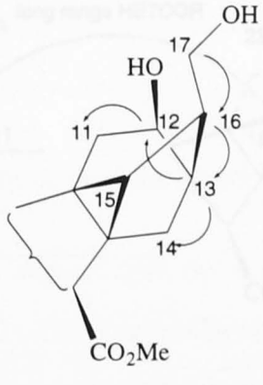


Figure 66. Important connectivities in the DQF COSY spectrum of <u>100</u>

The low intensity of the response correlating H(13) and H(12α), which is undoubtedly of crucial importance, can be explained in terms of the dihedral angle between these

two protons approaching 90°. Inspection of the Dreiding model suggested[91] that the size of the angle would place the value of the coupling constant in the range of 0.5-2 Hz. Moreover, the angle could be further increased towards 90° by changes in geometry caused by the steric repulsion between the C(16)-C(17)-O and C(11)-C(12)-O moieties (Figure 67).

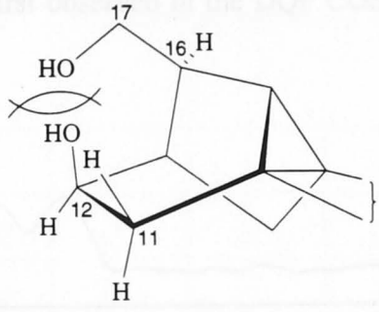


Figure 67. Steric interactions in the C/D ring region of 100

Additional evidence was therefore sought in order to confirm the correctness of these conclusions. A more global view of the environment of each assigned proton was acquired by the TOCSY spectrum with a mixing time of 40 milliseconds. No new information, however, was gained. The response from H(13) to H(12) was weak again as a result of slow magnetisation transfer caused by the small coupling. Two fragments, **A** and **B** were assembled on the base of the HMQC TOCSY[77,78] experiment (Figure 68).

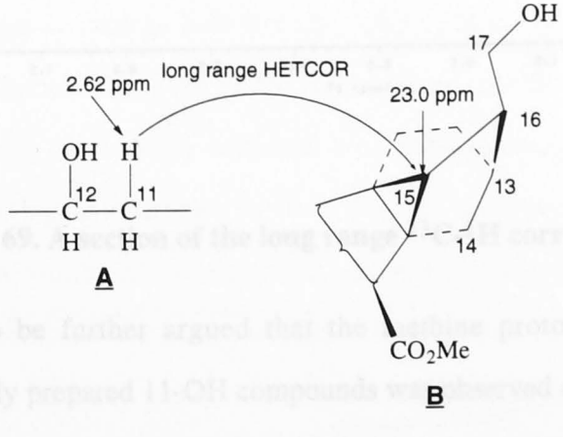


Figure 68. HMQC TOCSY experiment on 100

These fragments were joined using long a range HETCOR spectrum, as the cyclopropyl C(15) from fragment **B** exhibited a response to the methylene group proton

at 2.62 ppm in fragment **A**. Since long range heterocorrelated spectra of aliphatic molecules contain predominantly two- and three-bond correlations between heteroatoms[79] (a default value of 8 Hz for $^{n}J_{CH}$, where n = 2,3..., was used in the pulse sequence), the most sensible way of joining **A** and **B** was to assign the methylene group in **A** as C(11). It was thus confirmed that the low intensity of the crucial response between H(13) and H(12) first observed in the DQF COSY spectrum was indeed the result of a small coupling.

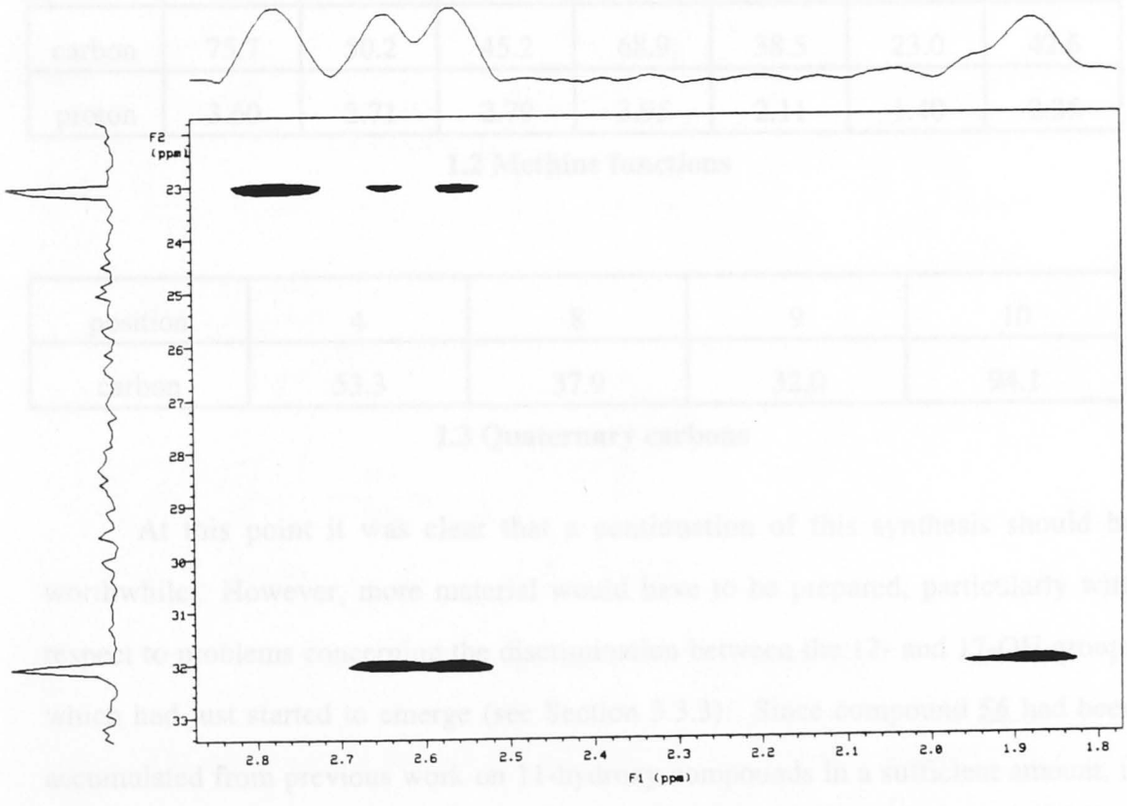


Figure 69. A section of the long range ^{13}C-^{1}H correlation

It could also be further argued that the methine proton of the -CH(OH)- fragment in previously prepared 11-OH compounds was observed at about 4.5-4.6 ppm, *i.e.* significantly downfield as compared to 3.95 ppm, because the protons at C(11) are deshielded by the cyclopropyl ring. Given all this evidence it could be concluded that compound **100** was the desired 12-OH derivative. Full assignment of the molecular framework was also made using the NMR experiments (Table 1).

Table 1. Assignment of skeletal atoms in 100 (chemical shifts are given in ppm)

position	1	2	11	14	17
carbon	24.4	24.8	27.5	32.1	62.7
Hα	1.89	1.64	2.62	1.60	3.85
Hβ	1.68	1.94	1.90	1.87	

1.1 Methylene functions

position	3	5	6	12	13	15	16
carbon	75.7	50.2	45.2	68.9	38.5	23.0	42.6
proton	3.60	2.71	2.79	3.95	2.11	1.40	2.35

1.2 Methine functions

position	4	8	9	10
carbon	53.3	37.9	32.0	94.1

1.3 Quaternary carbons

At this point it was clear that a continuation of this synthesis should be worthwhile. However, more material would have to be prepared, particularly with respect to problems concerning the discrimination between the 12- and 17-OH groups which had just started to emerge (see Section 3.3.3). Since compound **56** had been accumulated from previous work on 11-hydroxy compounds in a sufficient amount, it was decided to prepare the 3-deoxy series on a parallel basis. It was appreciated that this would mean having to characterize two series of compounds in future, but the potential advantages would be considerable. It would ensure that there would be enough material to complete the syntheses and the 3-deoxy compound **105** (Figure 70) would only have two hydroxy groups to be protected and deprotected. The 3-deoxy compounds would thus serve as a model series to select suitable procedures for elaborating the diol **100** towards the target molecules; an elimination protocol to reintroduce the 16-ene function in particular needed to be carefully chosen, owing to the sensitivity of the 9,15-cyclo-16-ene compounds.

Diol **105** was then constructed in a completely analogous manner to the preparation of **100** (Figure 70).

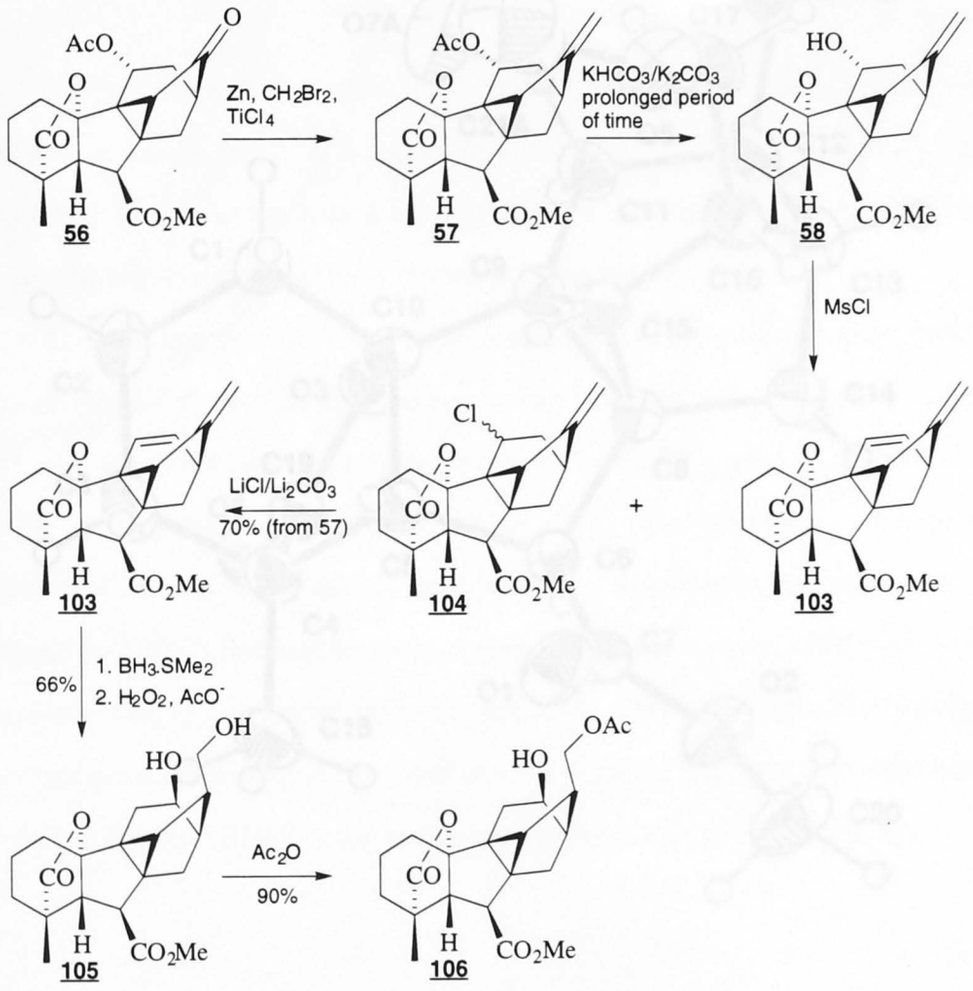
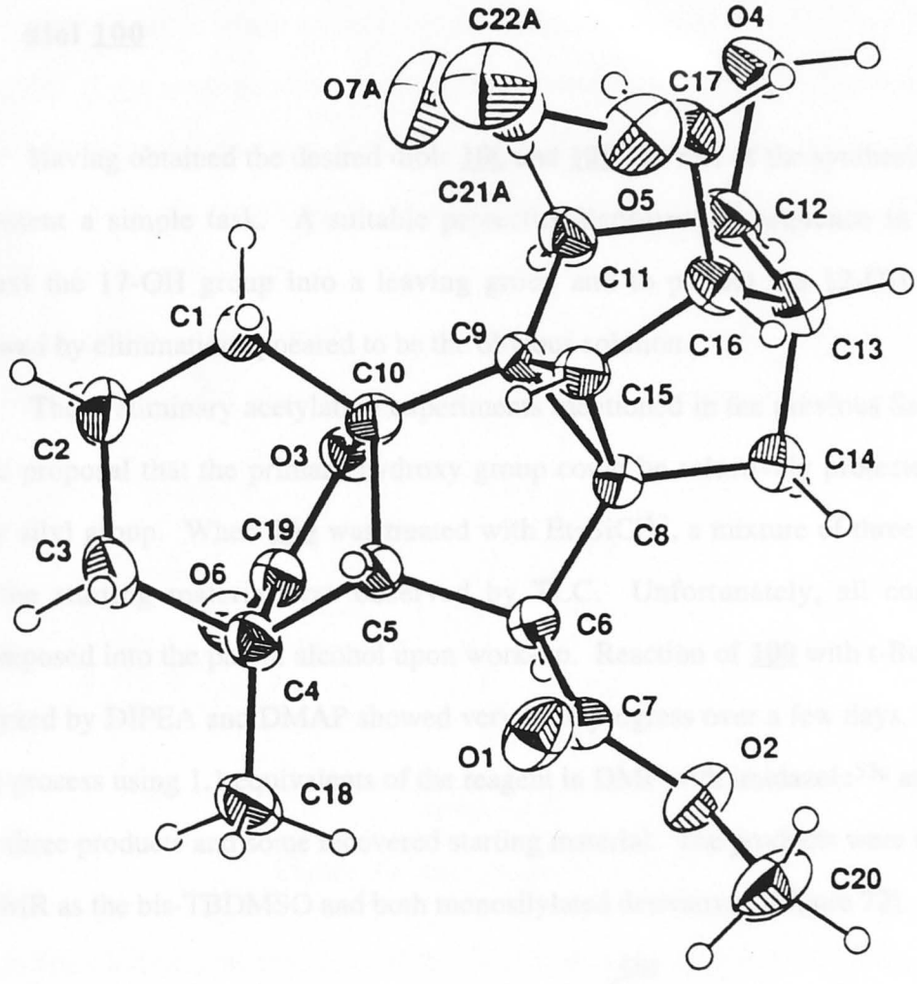
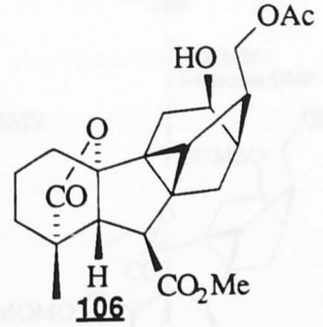


Figure 70. Preparation of 12β,17-diol 105

The acetylation of the diol **105** afforded monoacetate **106**, together with a small amount of the corresponding diacetate. Compound **106** crystallised from diethyl ether at -30°C to give discrete crystals, suitable for X-ray crystallography. X-ray analysis of the crystals (Figure 71) confirmed the location of both hydroxy groups, thus complementing the NMR studies carried out on the 3-substituted compound **100**. In summary, the formal transposition of the 11-substituent to C(12) was achieved in four steps from **95** and **58** to **100** and **105**, respectively.





106

Figure 71. Crystal structure of compound 106

3.3.3 Attempts to discriminate between the 12- and 17-OH groups in diol 100

Having obtained the desired diols **100** and **105**, the rest of the synthesis seemed to present a simple task. A suitable protection/deprotection sequence in order to convert the 17-OH group into a leaving group and to protect the 12-OH function followed by elimination appeared to be the obvious solution.

The preliminary acetylation experiments mentioned in the previous Section led to the proposal that the primary hydroxy group could be selectively protected with a bulky silyl group. When **100** was treated with Et_3SiCl[53], a mixture of three products and the starting material was observed by TLC. Unfortunately, all compounds decomposed into the parent alcohol upon work-up. Reaction of **100** with t-$BuMe_2SiCl$ catalyzed by DIPEA and DMAP showed very little progress over a few days, while the same process using 1.1 equivalents of the reagent in DMF with imidazole[53c] as the base gave three products and some recovered starting material. The products were identified by NMR as the bis-TBDMSO and both monosilylated derivatives (Figure 72).

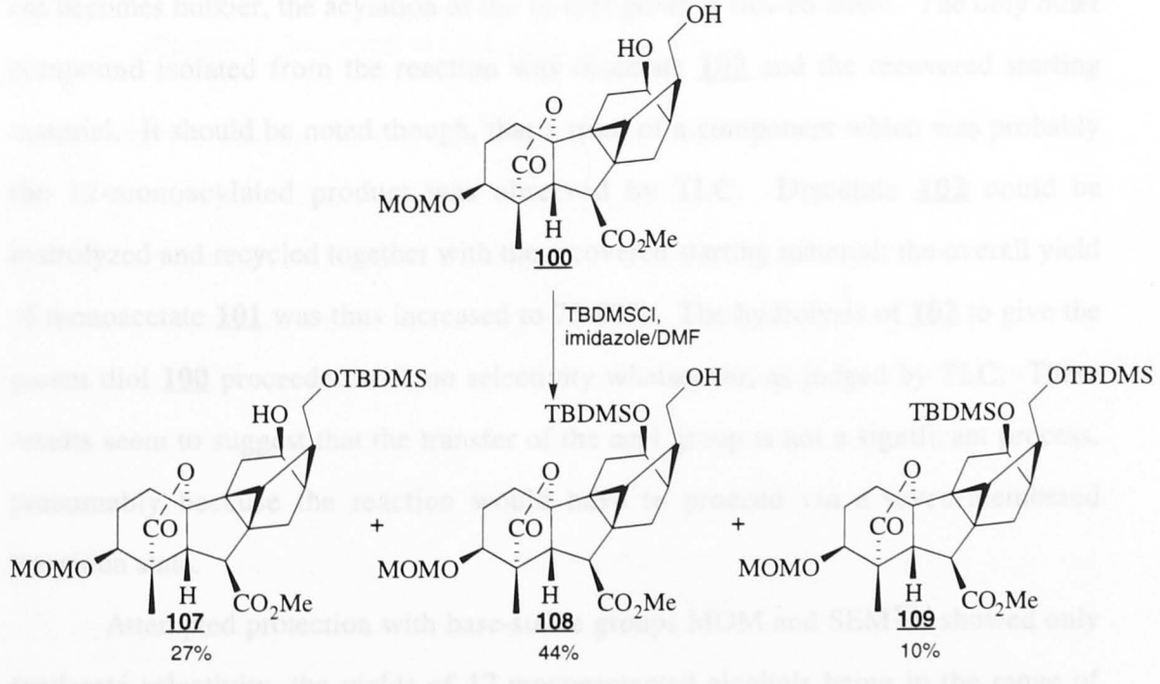


Figure 72. Attempted selective protection of diol 100 with TBDMSCl

These results indicated that the close spatial proximity of both OH groups had serious consequences which might endanger the completion of the synthesis. Given the selectivity of the corresponding acetylation (*vide infra*), it was concluded that after the initial silylation of the primary OH group the TBDMS moiety migrated to the 12-OH function. This is not surprising, as the migrations of silyl groups including TBDMS are described in literature[53c].

As already mentioned, acetylation was a reasonably selective process (Figure 73). Obviously, when the primary hydroxy group is esterified and the substitu-

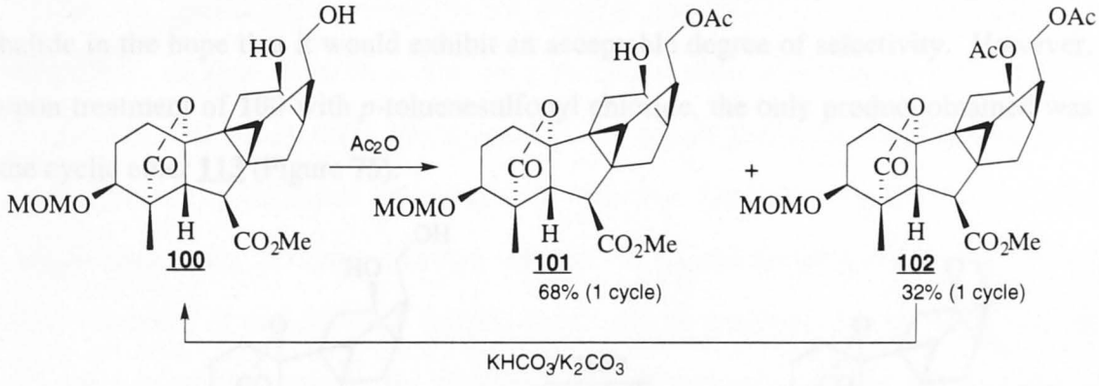


Figure 73. Acetylation of diol 100

ent becomes bulkier, the acylation of the 12-OH group is slowed down. The only other compound isolated from the reaction was diacetate **102** and the recovered starting material. It should be noted though, that a trace of a component which was probably the 12-monoacylated product was observed by TLC. Diacetate **102** could be hydrolyzed and recycled together with the recovered starting material; the overall yield of monoacetate **101** was thus increased to 75-80%. The hydrolysis of **102** to give the parent diol **100** proceeded with no selectivity whatsoever, as judged by TLC. These results seem to suggest that the transfer of the acyl group is not a significant process, presumably because the reaction would have to proceed *via* a seven-membered transition state.

Attempted protection with base-stable groups MOM and SEM[53d] showed only moderate selectivity, the yields of 17-monoprotected alcohols being in the range of 50-55% (Figure 74).

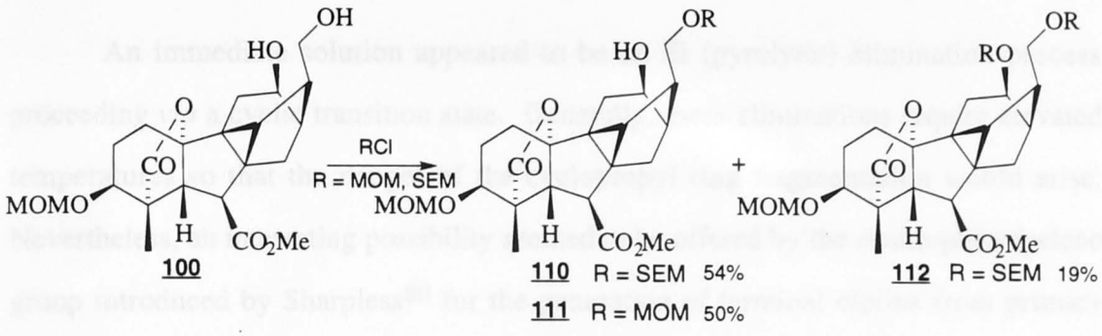


Figure 74. Protection of diol 100 with base-stable functions

Finally, given the objective of converting the 17-OH function into a leaving group, it was decided to treat diol **100** directly with a sterically demanding sulfonyl halide in the hope that it would exhibit an acceptable degree of selectivity. However, upon treatment of **100** with *p*-toluenesulfonyl chloride, the only product obtained was the cyclic ether **113** (Figure 75).

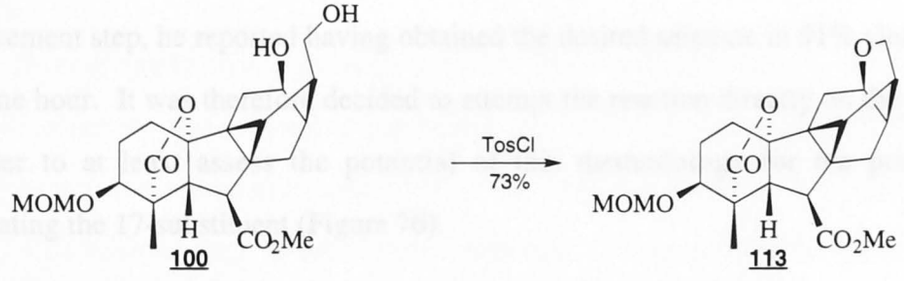


Figure 75. Reaction of diol 100 with p-toluenesulfonyl chloride

Clearly, once the primary hydroxy group is converted into a leaving group, the 12-OH function acts as an internal nucleophile, displacing the 17-tosyloxy function in an S_N2-like process, even under those mild conditions. This result showed that the successful hydroboration of dienes **96** and **103** was the beginning of yet another problem, the nature of which lay in the neighbouring group participation arising from the close spatial proximity of the two hydroxy groups. The 17-OH and 12β-OH functions are close to each other as a result of the stereochemical course of the hydroboration and, in this regard, the formation of **113** confirmed that the considerations on which this synthetic plan was based were correct. It could therefore be predicted at this point that any E1- or E2-type elimination of the 17-substituent, associated with the creation of a certain degree of positive charge at C(17) would be endangered in the same way.

An immediate solution appeared to be an Ei (pyrolytic) elimination process proceeding *via* a cyclic transition state. Generally, these eliminations require elevated temperatures so that the danger of the cyclopropyl ring fragmentation would arise. Nevertheless, an interesting possibility seemed to be offered by the *o*-nitrophenylseleno group introduced by Sharpless[80] for the generation of terminal olefins from primary alcohols. Primary hydroxy groups can be directly converted into the *o*-nitrophenylselenides with *o*-nitrophenylselenocyanate and Bu_3P in an S_N2 process[81] and the corresponding *o*-nitrophenylselenoxides can be eliminated under very mild conditions, furnishing terminal olefins in very good yields. Grieco published an example[81] of this transformation on a molecule with a relatively high level of complexity. Although there was a considerable potential for the neighbouring group participation by adjacent methoxy and acetoxy functions in the nucleophilic displacement step, he reported having obtained the desired selenide in 91% yield in less than one hour. It was therefore decided to attempt the reaction directly on the diol **100** in order to at least assess the potential of this methodology for the purpose of eliminating the 17-substituent (Figure 76).

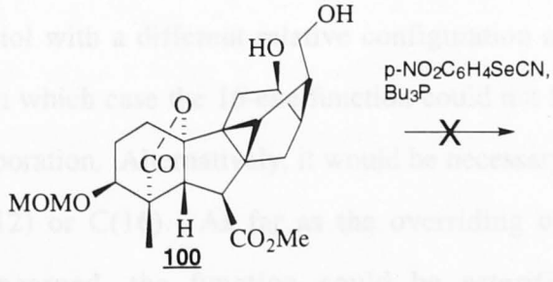


Figure 76. Attempted reaction of diol 100 with p-$NO_2C_6H_4SeCN$ and Bu_3P

The reagent was prepared according to the protocol of Bauer[80] and its quality confirmed by reaction with 1-dodecanol[80]. When compound **100** was subjected to the reaction conditions, the starting material was recovered after 10 hours at room temperature while a number of products were observed by TLC upon heating the reaction mixture. Literature examples[82] of the application of this procedure seem to indicate that the reaction may be sensitive to steric factors, due to the bulky nature of

both the nucleophile and the leaving group. Apparently, when the 17-OH group is activated by the phosphine, it assumes a position in which C(17) cannot be attacked by the RSe^- nucleophile along the trajectory required for an S_N2 process. Given the sterically crowded environment in the C/D ring region of **100**, the observed result is not surprising.

The need to tackle the problem arising from the neighbouring group participation was then obvious; attempted solutions are summarized in the next Section.

3.3.4 Attempts to deal with the neighbouring group participation

3.3.4.1 General considerations

Having found that a suitable substrate for the Ei elimination could not be prepared, attention was focused on preventing the 12-OH group in **100** (and, similarly, in **105**) from participating as a nucleophile in the processes leading to the reintroduction of the 16,17-double bond. In broad terms, two basic strategies could be used: either to avoid the participation or to override it. The former would mean modifying the synthesis so that a diol with a different relative configuration at C(12) and/or C(16) would be obtained, in which case the 16-ene function could not be used as the control element in the hydroboration. Alternatively, it would be necessary to invert the relative configuration at C(12) or C(16). As far as the overriding of the 12β-OH group participation is concerned, the function could be esterified with a strongly electron-withdrawing acyl moiety, such as trifluoroacetyl, in order to reduce the electron density on the oxygen. All approaches briefly considered here were attempted; the results are described in the following Sections.

3.3.4.2 Attempt to override the neighbouring group participation

As mentioned in the previous Section, the basic idea of this strategy was to acylate the 12-OH group in **100** with an acyl group with strongly electron-withdrawing substituents, such as Cl_3CCO or CF_3CO[83b]. Since these groups are extremely sensitive to basic media, the 17-OH moiety had to be protected with a function which could be removed under neutral or mild acidic conditions. Given the difficulties in protecting diol **100**, the choice of suitable protective groups was very limited. In the light of the results described in Section 3.3.3, it was decided to protect the 17-OH group as the trimethylsilylethoxymethyl ether, as it can be deprotected under a range of mild conditions[53d]. Regardless of the moderate selectivity of the protection procedure (see Section 3.3.3), it would allow the preparation of enough material for the subsequent reactions to be attempted and hence the feasibility of this approach could be assessed.

The 17-SEMO derivative **110** was treated with Cl_3CCOCl to afford the 12-trichloroacetoxy compound **114** (Figure 77). After initial experiments showed that $Bu_4N^+F^-$ was not a suitable reagent for the removal of the SEM group, it was found that CF_3COOH in dichloromethane at 0°C[53d] was satisfactory. However, this conversion could never be driven to completion and when the reaction was run for a prolonged time (more than 30-40 minutes), a number of byproducts were observed by both TLC and NMR. Nevertheless, enough compound **115** for the subsequent steps was accumulated.

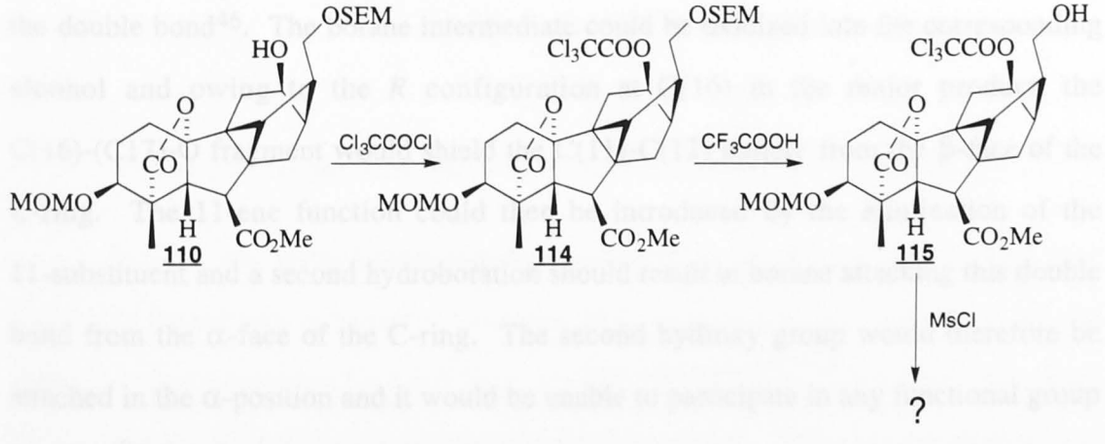


Figure 77. Attempt to override the neighbouring group participation

The crude alcohol **115** was treated with mesyl chloride in order to convert the 17-OH function into a leaving group. While TLC analysis revealed a number of products, one major product which was less polar than the starting material was isolated by column chromatography and attempts were made to elucidate its structure. NMR studies revealed the absence of the mesyloxy group resonance while the trichloroacetoxy function was still present, mass spectra being supportive of these conclusions. Unfortunately, the compound was contaminated with **114** due to the incomplete deprotection of the SEM group and more spectroscopic experiments would thus have been needed to determine the structure unequivocally. It could be concluded though, that the compound was not formed by a spontaneous elimination of MsOH, as the resonances characteristic of the 16-ene function were not located in the ^{1}H NMR spectrum. Given the low efficiency of each step starting from the protection of **100** with SEMCl, this approach was terminated in favour of another route, based on the inversion of configuration at C(16).

3.3.4.3 Attempt to perform two independent hydroborations

The basic considerations which led to the design of a slightly modified synthesis can be summarized as follows. If a hydroboration on a suitable 16-ene-11-substituted derivative was carried out, the borane would add predominantly from the exo-face of the double bond[46]. The borane intermediate could be oxidized into the corresponding alcohol and owing to the *R* configuration at C(16) in the major product, the C(16)-(C17)-O fragment would shield the C(11)-C(12) moiety from the β-face of the C-ring. The 11-ene function could then be introduced by the elimination of the 11-substituent and a second hydroboration should result in borane attacking this double bond from the α-face of the C-ring. The second hydroxy group would therefore be attached in the α-position and it would be unable to participate in any functional group interconversion reaction of the 17-OH group.

Compound **62** was the obvious candidate for the implementation of this plan and was therefore hydroborated with $BH_3.SMe_2$. Upon oxidative work-up, the reaction

afforded an inseparable mixture of 16-epimeric alcohols **116**, the desired 16 *R* epimer being the major one (16 *R* : 16 *S* = 5.5 : 1, as determined by ^{1}H NMR, Figure 78).

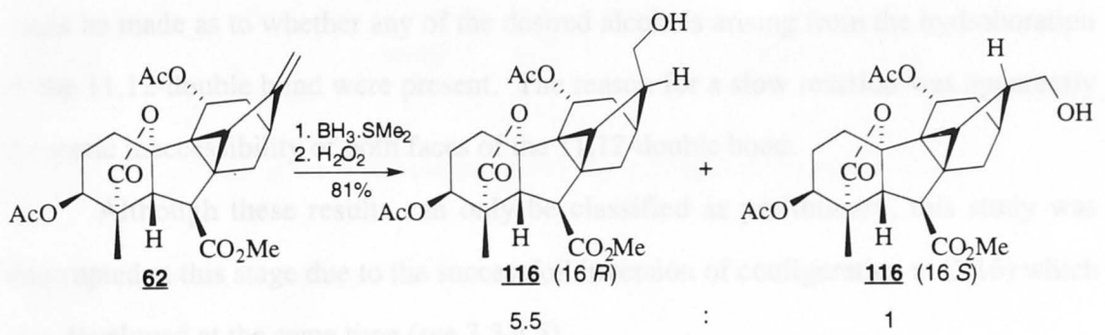


Figure 78. Hydroboration of the 16-ene 62

The primary hydroxy group was protected as the SEM-ether and the 3-acetoxy function was selectively hydrolyzed[54] and replaced with a MOM group. The resultant mixture of 16-epimers **117** was subjected to standard procedures for the elimination of the 11-substituent, described in Section 3.3.2. The 11-acetoxy group in **117** was removed with $KHCO_3/K_2CO_3$, the crude alcohol treated with mesyl chloride and the elimination completed with $LiCl/Li_2CO_3$ in DMF (Figure 79).

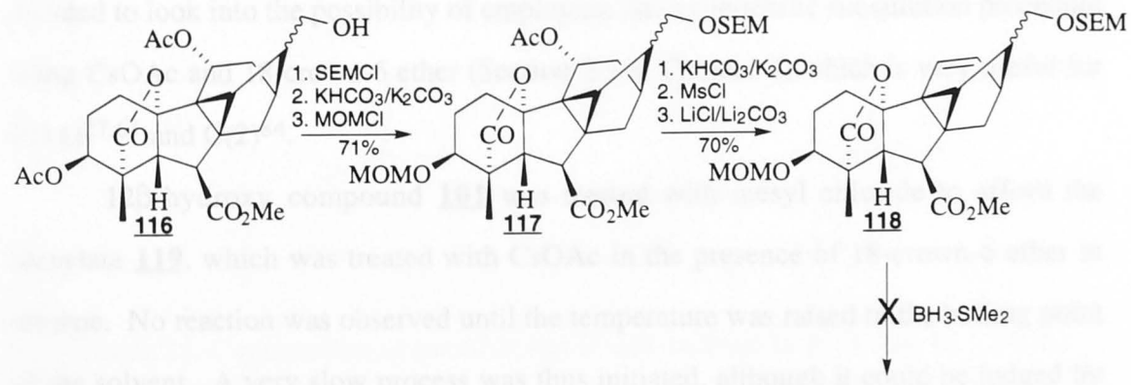


Figure 79. Preparation and attempted hydroboration of 11-enes 118

Finally, the same hydroboration conditions as for dienes **96** and **103** were applied to the mixture of isomeric 11-olefins **118**. Surprisingly, very little progress was observed after monitoring for a few hours by TLC; the addition of two more equivalents of the reagent did not significantly speed up the reaction. The reaction mixture was therefore heated at 40°C over 8 hours and although the starting material was still present by TLC, it was subjected to the usual oxidative work-up. TLC and NMR

analysis showed that a number of products were formed, among which a considerable amount of the 16 *R* epimer of the starting material was still detectable. No decision could be made as to whether any of the desired alcohols arising from the hydroboration of the 11,12-double bond were present. The reason for a slow reaction was apparently the steric inaccessibility of both faces of the 11,12-double bond.

Although these results can only be classified as preliminary, this study was interrupted at this stage due to the successfull inversion of configuration at C(16) which was developed at the same time (see 3.3.4.5).

3.3.4.4 Attempt to invert configuration at C(12)

Chu[44] attempted to implement a number of S_N2 processes, including the Mitsunobu protocol, in order to prepare 12α-OH gibberellins from their 12β-OH counterparts, the outcome being either a recovery of the starting material or decomposition. Regardless of the result being likely to be similar in this case, it was decided to look into the possibility of employing the nucleophilic substitution procedure using CsOAc and 18-crown-6 ether (Section 2.3.4, Chapter 2) which is very useful for C(11)[27,61] and C(2)[84].

12β-hydroxy compound **101** was treated with mesyl chloride to afford the mesylate **119**, which was treated with CsOAc in the presence of 18-crown-6 ether in toluene. No reaction was observed until the temperature was raised to the boiling point of the solvent. A very slow process was thus initiated, although it could be judged by the relative polarity of the main product that this was due to elimination rather than substitution. Indeed, ^{1}H NMR displayed signals at 6.24 and 5.75 ppm indicating the presence of the 11-ene function. Even after 3 days at reflux, a substantial amount of the starting material was still detected in the reaction mixture (Figure 80). The elimination can again be attributed to the steric congestion in the C-ring.

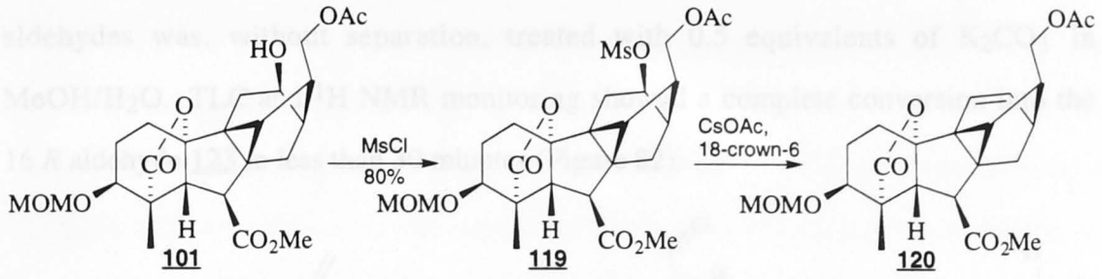


Figure 80. Attempt to invert the configuration at C(12)

3.3.4.5 Inversion of configuration at C(16)

The examination of a Dreiding model of diol **100** not only clarifies the problem of neighbouring group participation, but also suggests that, given the steric interactions between the C(16)-C(17)-O fragment and the β-substituents at C(11) and C(12), the 16 *R* configuration may be thermodynamically more favourable than the initially obtained 16 *S* (Figure 81). A question therefore arises regarding the possibility of inverting the absolute configuration at C(16) through, for example, enolization of a 17-aldehyde function.

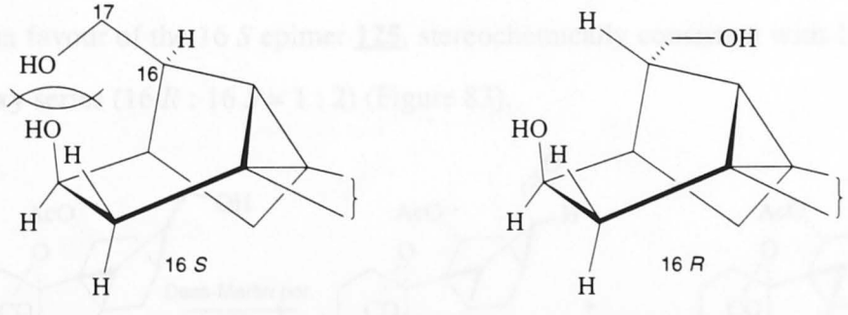


Figure 81. Comparison of possible steric interactions in 16 *R* and 16 *S* series

Brief model studies were carried out in order to probe the validity of these assumptions. 3-MOMO gibberellin A_4 methyl ester **121** available in the laboratory was treated with $BH_3.SMe_2$ followed by oxidative work-up to afford an inseparable mixture of 16-epimeric alcohols, which were oxidized with the Dess-Martin periodinane[64] into the corresponding aldehydes **122** and **123**. The ratio of **122** to **123** was found to be 2 : 1 by ^1H NMR, the 16 *S* configuration (corresponding to 16 *S* in the 12-hydroxy-9,15-cyclo compounds) of the major aldehyde being again the consequence of the predominant exo-addition of borane across the 16,17-double bond. The mixture of

aldehydes was, without separation, treated with 0.5 equivalents of K_2CO_3 in $MeOH/H_2O$. TLC and ^1H NMR monitoring showed a complete conversion into the 16 *R* aldehyde **123** in less than 30 minutes (Figure 82).

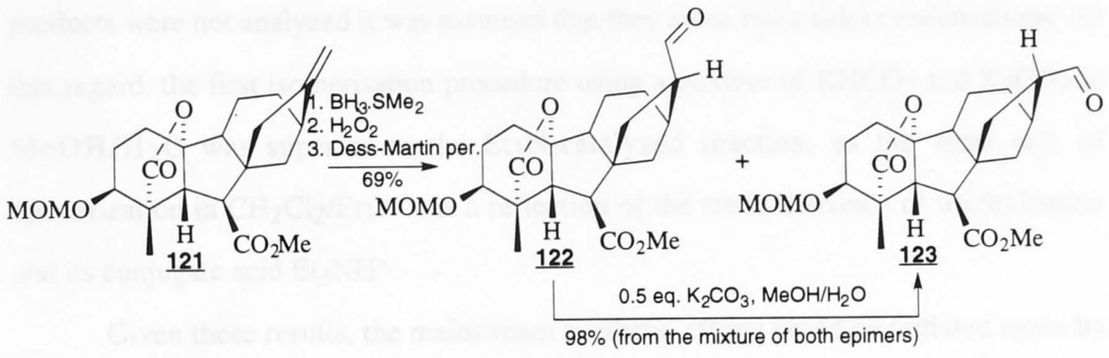


Figure 82. Inversion of configuration at C(16) in the model series

This outcome was very encouraging and prompted further studies on the 9,15-cyclo system. Mixture **116** (see Figure 78) was oxidized into the corresponding aldehydes (**124** and **125**), which could not be distinguished by TLC. Since the molecule contained two acetoxy functions, another base, triethylamine, was used to promote epimerization at C(16) in CH_2Cl_2 as the solvent. As expected, the original ratio changed in favour of the 16 *S* epimer **125**, stereochemically consistent with 16 *R* in the 12-hydroxy series (16 *R* : 16 *S* = 1 : 2) (Figure 83).

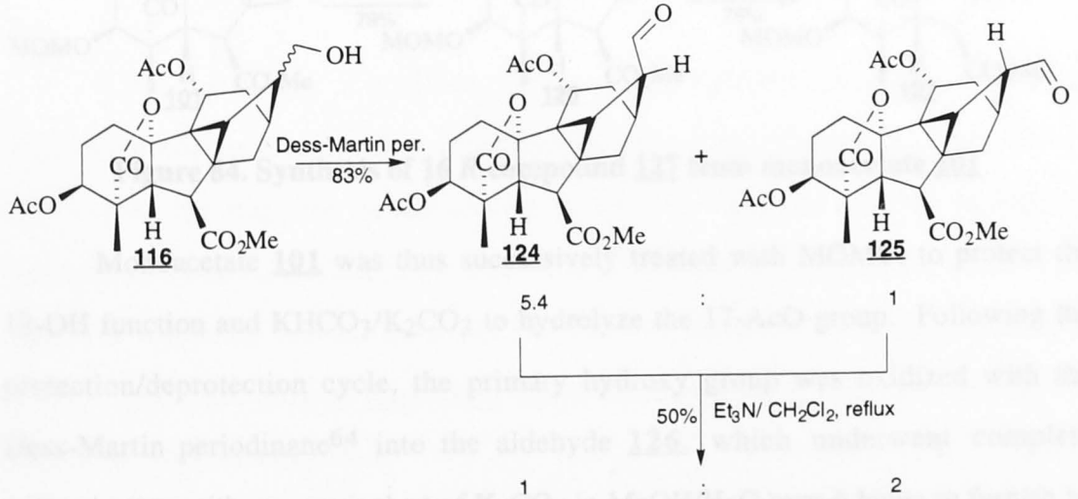


Figure 83. C(16) epimerization in the 9,15-cyclo system

The moderate yield of this reaction could be ascribed to the reaction conditions (12 hours at reflux, CH_2Cl_2/Et_3N, 2 : 1). It was found that all 17-aldehydes which were

prepared for the purpose of these studies did not tolerate a long exposure to strongly basic and acidic media and that they could not be stored for a long time. Conversions into markedly more polar compounds were observed by TLC and although these products were not analyzed it was assumed that they arose from aldol condensations. In this regard, the first isomerisation procedure using a mixture of $KHCO_3$ and K_2CO_3 in $MeOH/H_2O$ was superior to the Et_3N-catalyzed reaction, as the slow rate of epimerization in CH_2Cl_2/Et_3N was a reflection of the steric demands of triethylamine and its conjugate acid Et_3NH^+.

Given these results, the mainstream synthetic efforts could be initiated again by applying the procedure to a suitable 12-protected-17-oxo compound derived from diol **100**. Monoacetate **101** was selected as the starting material due to the high yield in which it could be obtained and for the ease with which the 17-acetoxy group could be removed. The first step was therefore the protection of the 12-hydroxy group with a base-stable protective function in order to allow selective oxidation of the primary hydroxy group.

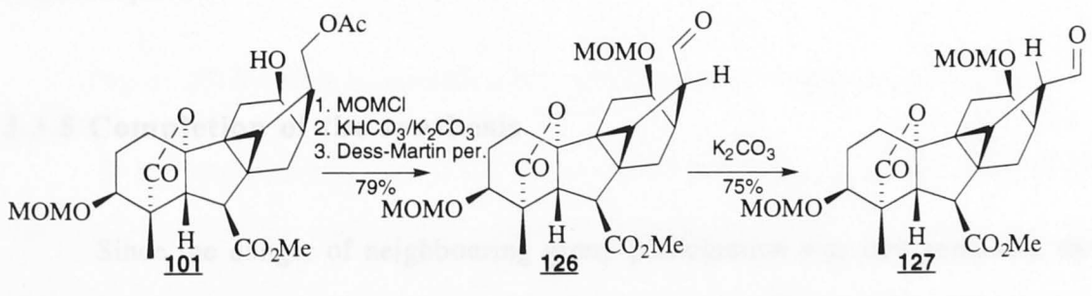


Figure 84. Synthesis of 16 *R* compound 127 from monoacetate 101

Monoacetate **101** was thus successively treated with MOMCl to protect the 12-OH function and $KHCO_3/K_2CO_3$ to hydrolyze the 17-AcO group. Following the protection/deprotection cycle, the primary hydroxy group was oxidized with the Dess-Martin periodinane[64] into the aldehyde **126**, which underwent complete epimerization with one equivalent of K_2CO_3 in $MeOH/H_2O$ over 6 hours to furnish its 16 *R* counterpart **127** in good yield. Since the epimeric aldehydes could not be distinguished by TLC in a wide variety of solvent systems, the progress of the reaction could only be checked by ^1H NMR. The NMR monitoring was based on the aldehyde

proton resonance in **126** at 9.92 ppm and the corresponding signal from **127** at 9.67 ppm.

Similarly, the same sequence of reactions was applied to the 3-deoxy compound **106** (Figure 85); in contrast to **126** and **127** , aldehydes **128** and **129** were observed as two spots of different R_f values by TLC.

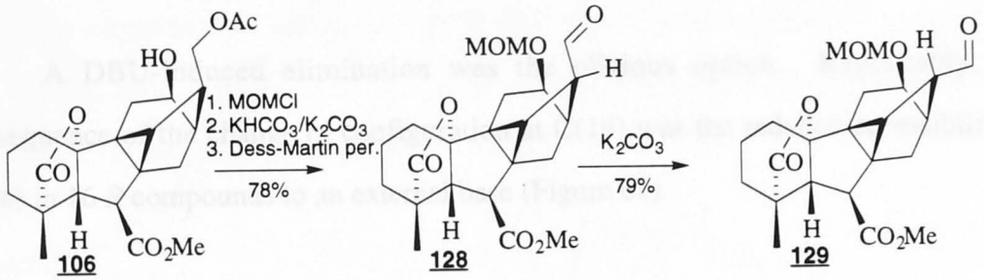


Figure 85. Synthesis of the 3-deoxy compound 129

With aldehydes **127** and **129** in hand, the synthesis of the desired targets could be completed by elimination as the last synthetic operation. Unfortunately, the necessity to invert the relative configuration at C(16) added a few extra steps to the overall sequence.

3.3.5 Completion of the synthesis

Since the danger of neighbouring group participation was now removed, the sequence could be completed in a straightforward manner (Figure 86), commencing with setting the stage for the elimination of the 17-substituent. Aldehyde **129** was reduced with $NaBH_4$ and the crude alcohol treated[85] with Ph_3P/I_2 to afford the iodide **130**. The secondary hydroxy group at C(12) was then liberated[86] with Me_2BBr to yield alcohol **131**. Elimination procedures could subsequently be employed in order to restore the 16,17-double bond.

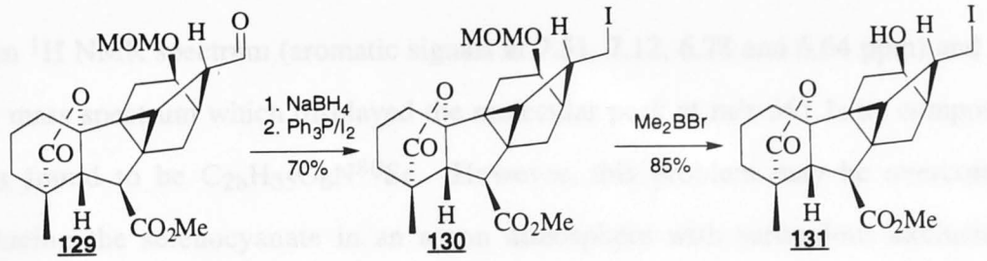


Figure 86. Conversion of the 17-oxo function into an iodo moiety

A DBU-induced elimination was the obvious option. Regrettably, one consequence of the change of configuration at C(16) was the reduced accessibility of H(16) in 16 *R* compounds to an external base (Figure 87).

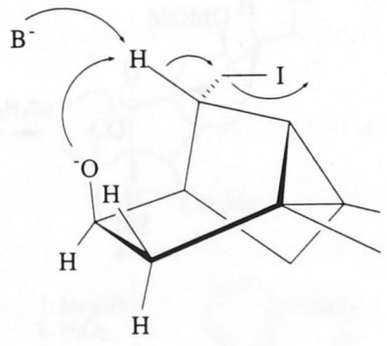


Figure 87. Possible competition between the external and internal base

It was hoped though, that if the 12-OH group was unprotected, the anion of the alcohol formed during the reaction could act as an internal base, thus facilitating the process.

With a view to having alternative means of effecting the elimination process, another protocol based on the Ei mechanism was implemented on a parallel basis. Iodide **130** was thus treated with a solution of *o*-nitrophenylselenide anion generated by the reduction of *o*-nitrophenylselenocyanate with sodium borohydride[80]. The substitution afforded two compounds, the major product being the desired *o*-nitrophenylselenide **132**, as indicated by the signals of the aromatic protons in the ^{1}H NMR spectrum (8.3, 7.51 and 7.83 ppm). The other compound was identified as the *o*-aminophenylselenide **133**, the formation of which can be explained by the reduction of the aromatic nitro group by an excess of $NaBH_4$. The structure of **133** was clear

from ^{1}H NMR spectrum (aromatic signals at 7.51, 7.12, 6.78 and 6.64 ppm) and from the mass spectrum which displayed the molecular peak at m/z 561.1; its composition was found to be $C_{28}H_{35}O_6N^{80}Se$. However, this problem may be overcome by reducing the selenocyanate in an argon atmosphere with scrupulous exclusion of atmospheric oxygen which otherwise participates in the oxidation of the *o*-nitrophenylselenide anions[80]. It is then possible to control the stoichiometry accurately, instead of using an excess of sodium borohydride in order to reduce the diselenide formed during the reaction.

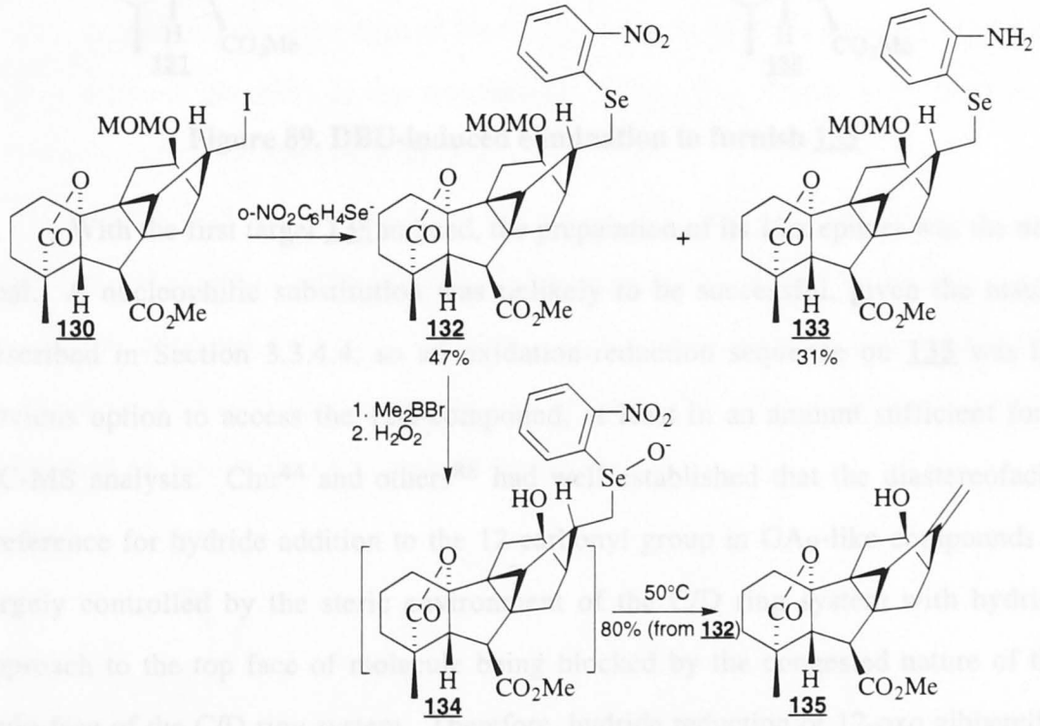


Figure 88. Selenoxide elimination protocol for the preparation of 135

Selenide **132** was oxidized with hydrogen peroxide to yield the selenoxide **134**, which turned out to be remarkably stable. Only the starting material was detected by TLC after 15 hours at room temperature while not even a sign of the formation of olefinic products was observed. Nevertheless, addition of triethylamine[87] and heating at 50°C initiated a somewhat slow Ei elimination to furnish the desired compound **135**.

Attempting the alternative DBU-induced elimination procedure, iodide **131** was treated with DBU in DMF to give rise to the same olefin **135** (Figure 89). TLC monitoring showed that the elimination process was very slow, the reaction requiring

three days at 50°C to proceed to completion. The slow reaction rate was attributed to H(16) being in a sterically hindered environment for the bulky base DBU (Figure 87); obviously, the 12-OH group did not provide any assistance whatsoever. The structure of **135** was confirmed by ^{1}H NMR (singlets at 5.01 and 4.90 ppm for the exocyclic methylene group) and the APT spectrum.

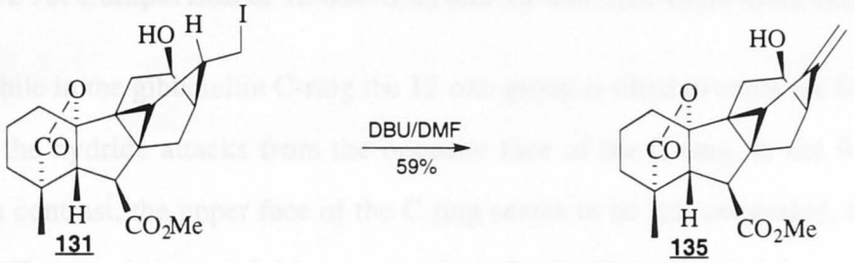


Figure 89. DBU-induced elimination to furnish 135

With the first target **135** in hand, the preparation of its 12α epimer was the next goal. A nucleophilic substitution was unlikely to be successful, given the results described in Section 3.3.4.4, so an oxidation-reduction sequence on **135** was the obvious option to access the 12α-compound, at least in an amount sufficient for a GC-MS analysis. Chu[44] and others[88] had well established that the diastereofacial preference for hydride addition to the 12-carbonyl group in GA_9-like compounds is largely controlled by the steric environment of the C/D ring system with hydride approach to the top face of molecule being blocked by the congested nature of the endo-face of the C/D ring system. Therefore, hydride reduction of 12-oxo gibberellin derivatives normally returns the 12β-alcohol as the sole product (Figure 90). The diastereoselectivity of the reduction can only be reversed if the 12-keto function is chelated with the 13-OH group, as the chelation changes the conformation of the C-ring from a boat to a half-chair[43,44]. However, the inspection of a Dreiding model suggested that the 9,15-cyclo system is slightly different from that of GA_9-like gibberellins.

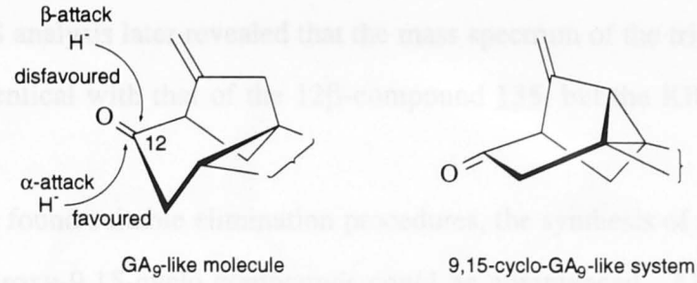


Figure 90. Comparison of 12-oxo-GA$_9$ and 12-oxo-9,15-cyclo-GA$_9$ systems

While in the gibberellin C-ring the 12-oxo group is tilted towards the D-ring and therefore the hydride attacks from the opposite face of the C-ring, in the 9,15-cyclo system, in contrast, the upper face of the C ring seems to be less congested, due to the slightly different geometry of this arrangement. It was thus hoped that at least some 12α-alcohol would be obtained upon reduction.

Alcohol **135** was oxidized into the ketone **136**, the reduction of which with $NaBH_4$ at -60°C returned mainly the starting material (Figure 91), but the 12α-epimer was clearly observed by TLC. The same process carried out at room temperature produced an increased amount of the 12α-alcohol **137**. The actual ratio was not determined because of the small scale of the reaction.

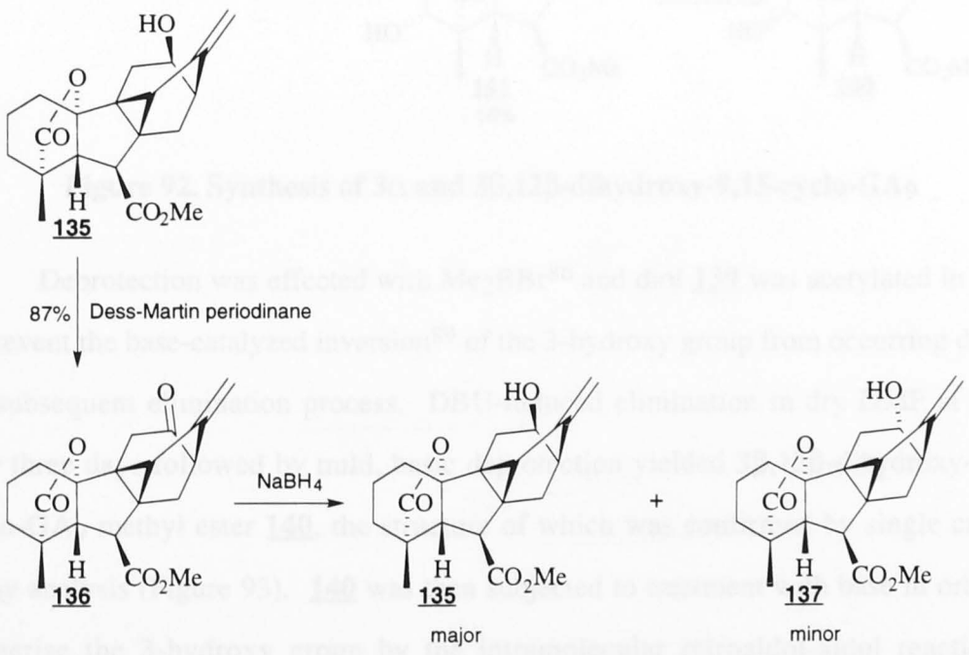


Figure 91. Oxidation/reduction sequence on 135

GC-MS analysis later revealed that the mass spectrum of the trimethylsilyl ether of **137** was identical with that of the 12β-compound **135**, but the KRI differed by 30 units.

Having found suitable elimination procedures, the synthesis of the 12β-series of the 3,12-dihydroxy-9,15-cyclo compounds could be commenced. Aldehyde **127** was reduced and the crude alcohol converted[85] into iodide **138** with Ph_3P/I_2 (Figure 92). The MOM groups had to be removed at this stage because of the already mentioned sensitivity of the 16-ene function towards acidic conditions.

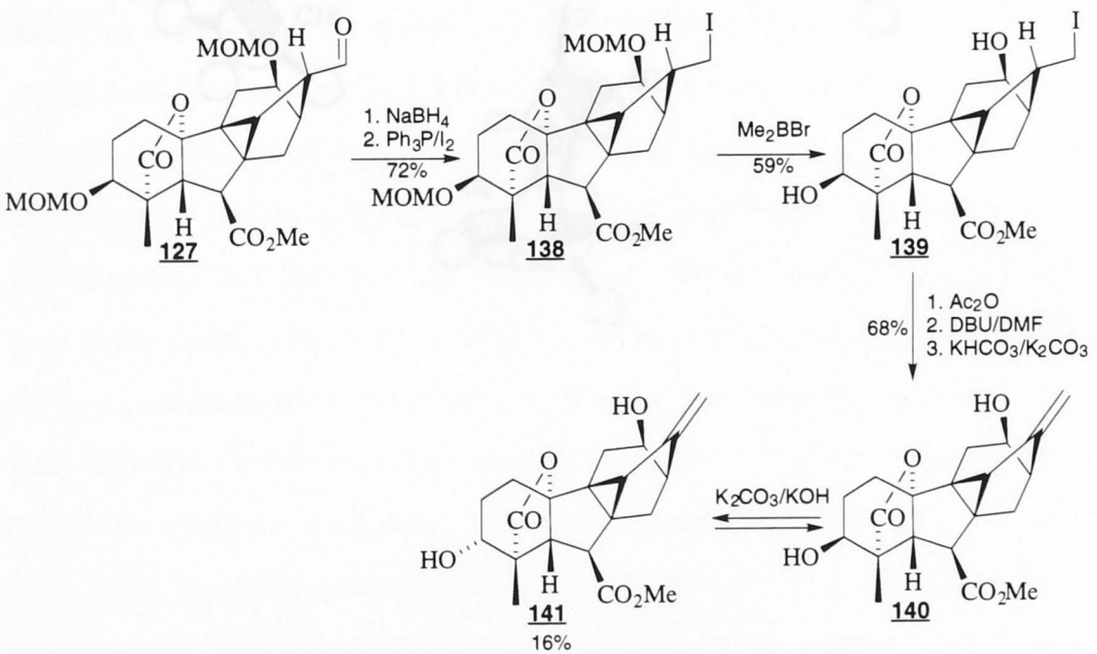
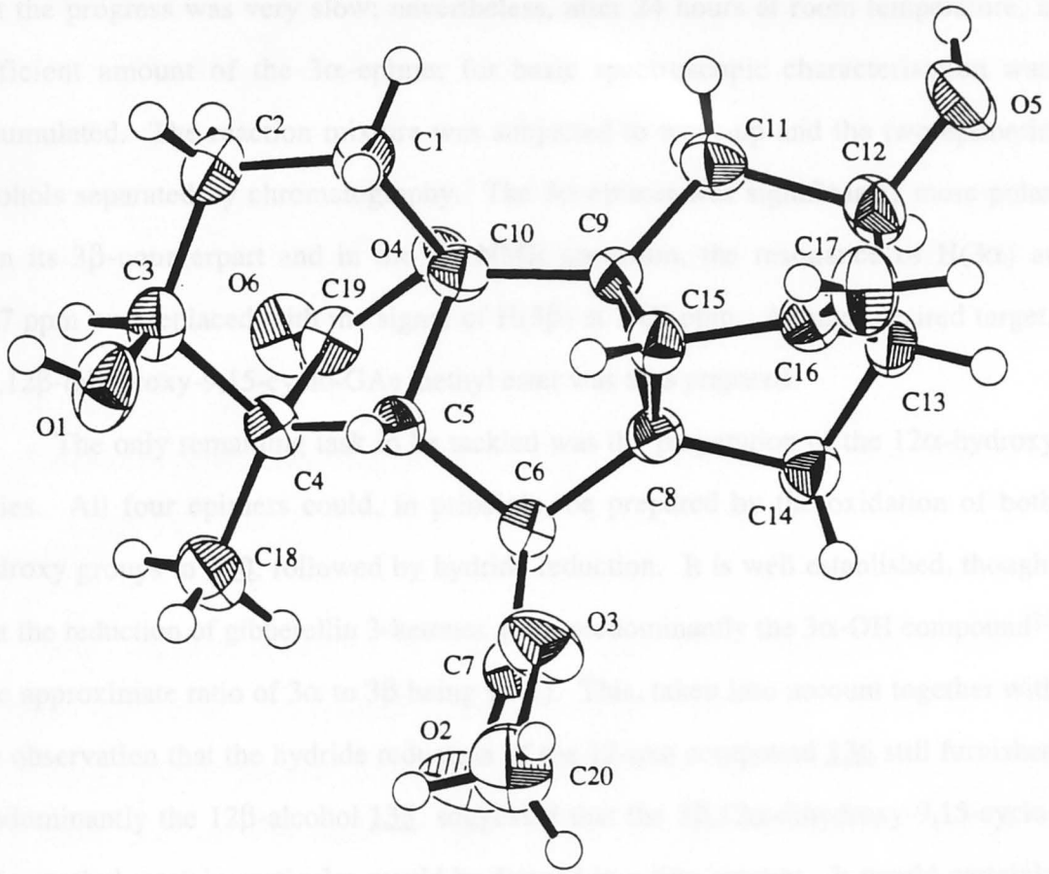
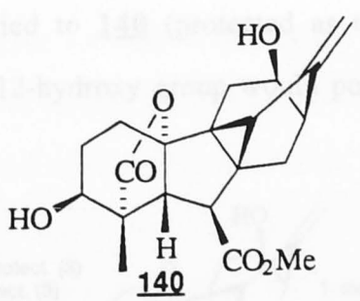


Figure 92. Synthesis of 3α and 3β,12β-dihydroxy-9,15-cyclo-GA_9

Deprotection was effected with Me_2BBr[86] and diol **139** was acetylated in order to prevent the base-catalyzed inversion[89] of the 3-hydroxy group from occurring during the subsequent elimination process. DBU-induced elimination in dry DMF at 50°C over three days followed by mild, basic deprotection yielded 3β,12β-dihydroxy-9,15-cyclo-GA_9 methyl ester **140**, the structure of which was confirmed by single crystal X-ray analysis (Figure 93). **140** was then subjected to treatment with base in order to epimerise the 3-hydroxy group by the intramolecular retroaldol-aldol reaction[89], thereby providing access to the 3α,12β-dihydroxy isomer. TLC monitoring showed





140

Figure 93. Crystal structure of compound 140

that the progress was very slow; nevertheless, after 24 hours at room temperature, a sufficient amount of the 3α-epimer for basic spectroscopic characterisation was accumulated. The reaction mixture was subjected to work-up and the two epimeric alcohols separated by chromatography. The 3α-epimer was significantly more polar than its 3β-counterpart and in the ^{1}H NMR spectrum, the resonance of H(3α) at 3.87 ppm was replaced with the signal of H(3β) at 3.60 ppm. Another desired target, 3α,12β-dihydroxy-9,15-cyclo-GA$_9$ methyl ester was thus prepared.

The only remaining task to be tackled was the preparation of the 12α-hydroxy series. All four epimers could, in principle, be prepared by the oxidation of both hydroxy groups in **140**, followed by hydride reduction. It is well established, though, that the reduction of gibberellin 3-ketones gives predominantly the 3α-OH compound[20] (the approximate ratio of 3α to 3β being 9 : 1). This, taken into account together with the observation that the hydride reduction of the 12-oxo compound **136** still furnished predominantly the 12β-alcohol **135**, suggested that the 3β,12α-dihydroxy-9,15-cyclo-GA$_9$ methyl ester in particular would be formed in a tiny amount. It would certainly not be a problem in terms of GC-MS analysis, but it was desirable to obtain at least basic ^{1}H NMR data on each diastereomer. An alternative option was to utilize an observation which was made during the preparation of the 3β,12β-dihydroxy derivative **140**. When the elimination was complete and the protected target being hydrolyzed (see Figure 92), TLC monitoring showed that one of the acetoxy groups (most likely the 3-AcO function[54]) was hydrolyzed faster than the other. Therefore, a protection/ deprotection sequence applied to **140** (protected as the diacetate), followed by oxidation/ reduction of the 12-hydroxy group would potentially provide a solution (Figure 94).

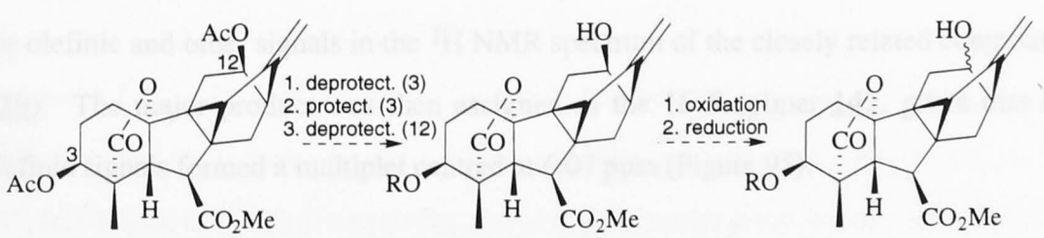


Figure 94. Possible route to the 12α-series

The synthesis would be completed, again, by base-catalyzed inversion of the 3β-OH group[89] (*vide supra*). However, this plan could not be implemented because of the lack of material.

It was fortunate then, that a 3β,12α-di-OH compound was discovered upon examining the byproducts from the hydroboration of diene **96**. The results of complete structure elucidation on these compounds are described in the next Section.

3.3.6 Structure elucidation of byproducts from the hydroboration of diene 96

As mentioned in Section 3.3.2 of this Chapter, the hydroboration of diene **96** afforded two minor gibberellin products (**A** and **B**). Since they accounted for a relatively significant percentage of the total mass of the products formed in the reaction, an obvious decision was made to carry out spectroscopic experiments in order to elucidate their structures. Moreover, a full picture of the hydroboration reaction would thus be obtained.

A one-dimensional proton spectrum provided enough information about the structure of product **A**, which was less polar than the main product, diol **100**. **A**, which appeared as one spot by TLC was found to be a mixture of two compounds. Olefinic signals restricted to the range 5.7 to 6.3 ppm clearly showed that both derivatives had lost the 16-ene function, but still possessed the 11,12-double bond and must therefore have originated from the simple addition of $BH_3.SMe_2$ across the exocyclic double bond. The olefinic resonances of the minor compound at 6.23 ppm (a doublet) and 5.78 ppm (a doublet of doublets) were consistent with the 16 *R* derivative **143** (based on the olefinic and other signals in the ^1H NMR spectrum of the closely related compound **120**). The major product was then assigned as the 16 *S* epimer **142**, given that its olefinic signals formed a multiplet centred at 6.07 ppm (Figure 95).

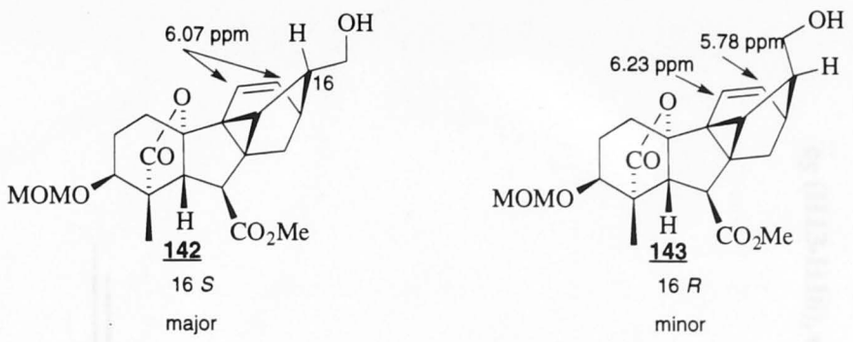


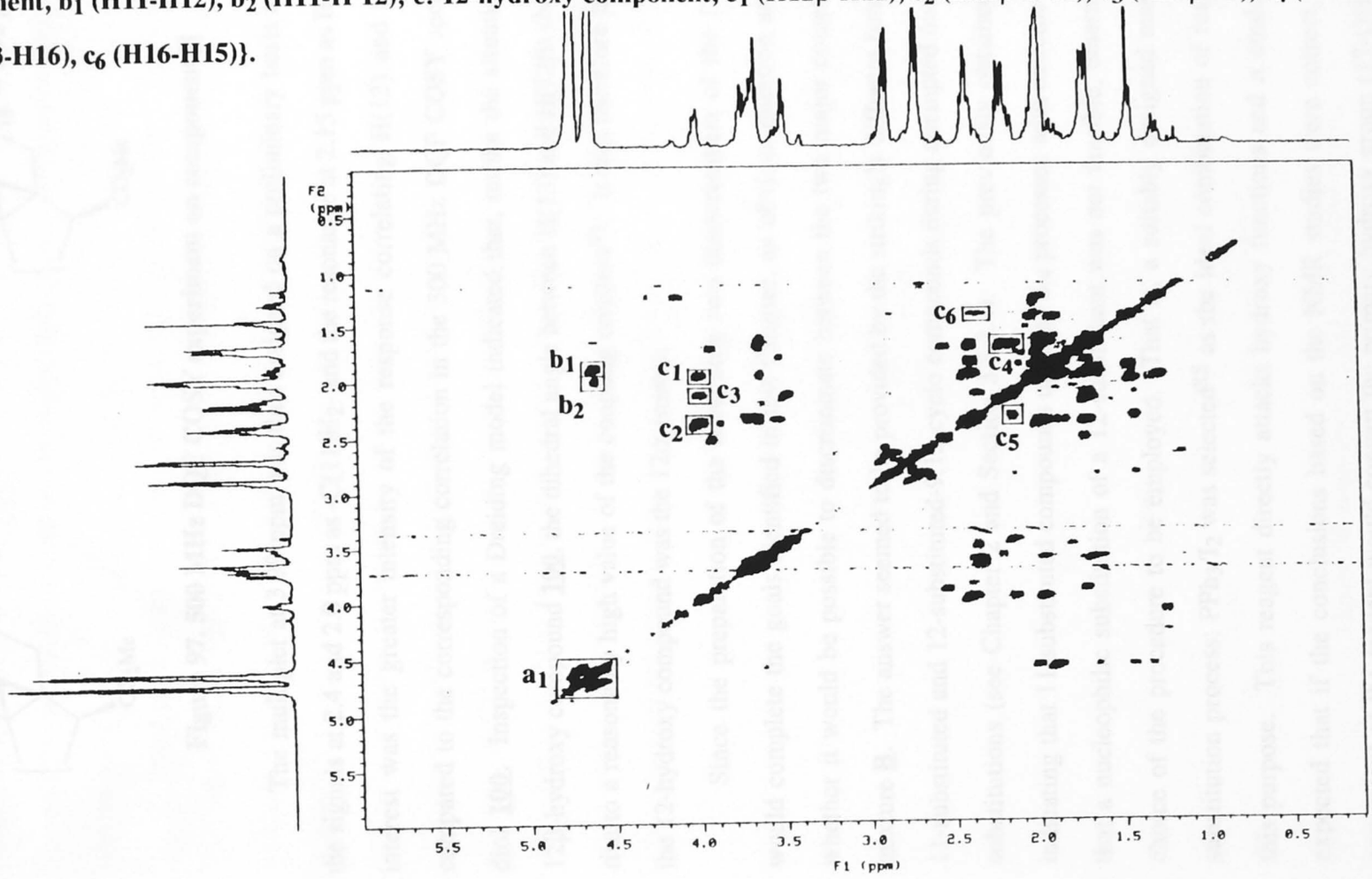
Figure 95. Composition of the hydroboration byproduct A

More interesting was product **B**, which was slightly more polar than diol **100**. It could be judged from its relative polarity that it was a diol or a mixture of diols. The ^1H NMR spectrum indicated that it was indeed a mixture of two major compounds and possibly a few minor components. The presence of a multiplet at 3.99 ppm suggested that one of the components may have been a 12-hydroxy compound.

A 500 MHz DQF COSY experiment[76] (Figure 96) provided further insight into the structures. If one of the compounds was an 11-hydroxy derivative, the signal of H(11) would appear in the range of 4.5-4.7 ppm and it would therefore be overlapped by the resonance of the methylene protons of the MOM group protecting the 3-hydroxy function. In the 2D spectrum, the AB system of the $-OCH_2O-$ moiety at 4.67 ppm was clearly identified as two diagonal and two close-to-diagonal responses. Two additional off-diagonal correlations which could not arise from the $-OCH_2O-$ moiety were observed at the same chemical shift. They correlated a signal, which was overlapped by the $-OCH_2O-$ resonance, to protons at 1.87 and 1.99 ppm, which, in turn, were strongly coupled to each other, displaying an AB-like pattern. Since this would be expected for a -CH(OH)- group with neighbouring quaternary and methylene carbons, the presence of an 11-hydroxy compound was a distinct possibility.

Three responses were recorded for the multiplet at 3.99 ppm (*vide supra*), to protons at 2.40, 2.15 and 2.00 ppm. The resonances at 2.40 and 2.00 ppm correlated to each other and the pattern was again diagnostic of the AB system of a methylene group. The appearance of the third correlation confirmed the initial suspicion that a 12-hydroxy compound was one of the major components in the mixture (Figure 97).

Figure 96. 500 MHz DQF COSY spectrum of mixture B {selected responses: a_1 (-OCH_2O- in -OMOM); b: 11-hydroxy component, b_1 (H11-H12), b_2 (H11-H'12); c: 12-hydroxy component, c_1 (H12β-H11), c_2 (H12β-H'11), c_3 (H12β-13), c_4 (H13-H14β), c_5 (H13-H16), c_6 (H16-H15)}.



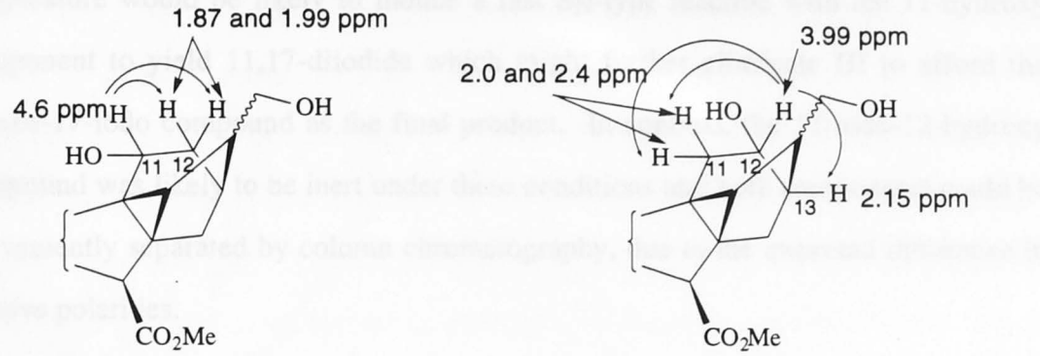


Figure 97. 500 MHz DQF COSY experiment on component B

The multiplet at 3.99 ppm was thus assigned on a preliminary basis as H(12), the signals at 2.4 and 2.0 ppm as -C(11)H_2- and the resonance at 2.15 ppm as H(13). Of interest was the greater intensity of the response correlating H(12) and H(13) as compared to the corresponding correlation in the 500 MHz DQF COSY spectrum of diol **100**. Inspection of a Dreiding model indicated that, unlike the situation in the 12β-hydroxy compound **100**, the dihedral angle between H(13) and H(12β) should give rise to a reasonably high value of the coupling constant[91]. It was therefore likely that the 12-hydroxy compound was the 12α-isomer.

Since the preparation of the remaining two diastereomers of the 12α-series would complete the goals identified in this Chapter, an obvious question arose as to whether it would be possible to discriminate between the two major components in mixture **B**. The answer seemed to be provided by the strikingly different behaviour of 11-substituted and 12-substituted-9,15-cyclo compounds during attempted nucleophilic substitutions (see Chapter 2 and Section 3.3.3.4). The previously obtained results, indicating that 11-substituted compounds undergo S_N processes with extreme ease and that a nucleophilic substitution of a 12-substituent was not feasible, determined the choice of the procedure to be employed. That is, a suitably designed nucleophilic substitution process; PPh_3/I_2 was selected[85] as the ideal combination of reagents for this purpose. This reagent directly attacks hydroxy functions and it could well be expected that if the conclusions based on the NMR studies were correct, the first equivalent of the reagent would convert the primary hydroxy group (17-OH) in each derivative into an iodo function. A second equivalent of PPh_3/I_2 added at room

temperature would be likely to induce a fast S_N-type reaction with the 11-hydroxy component to yield 11,17-diiodide which might further eliminate HI to afford the 11-ene-17-iodo compound as the final product. In contrast, the 17-iodo-12-hydroxy compound was likely to be inert under these conditions and both components could be conveniently separated by column chromatography, due to the expected difference in relative polarities.

These predictions turned out to be correct. A rapid reaction was observed by TLC when the first portion (1.3 eq.) of the reagent was added, whereby the starting material was converted into a mixture of two products of very similar R_f values. Another 1.3 equivalents of PPh_3/I_2 reacted with one of the compounds to give a relatively non-polar product of a high R_f value, leaving the other one intact. ^1H NMR experiments identified the non-polar compound as a mixture of 17-iodo-11-enes **146**, epimeric at C(16) (Figure 98), based on olefinic signals at 5.80 ppm (a triplet) and 6.32 ppm (a doublet) for the 16 *R* isomer and a multiplet at 6.04 ppm for the 16 *S* compound. Unfortunately, owing to the elimination of HI from the C-ring the stereochemical information about the configuration of the 11-hydroxy group was lost.

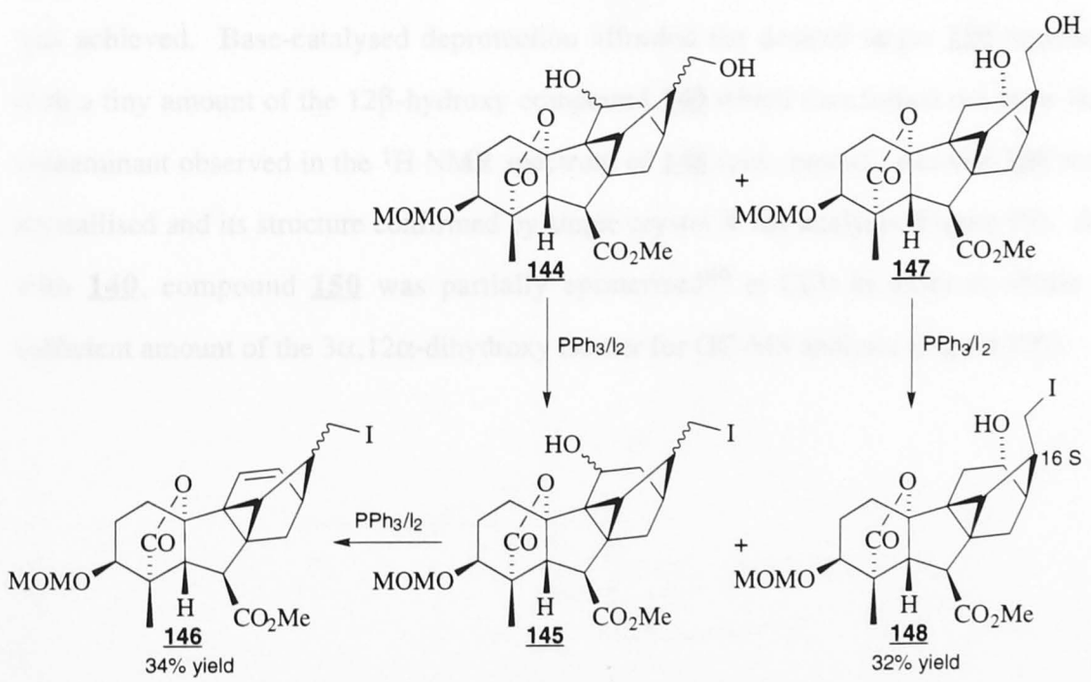
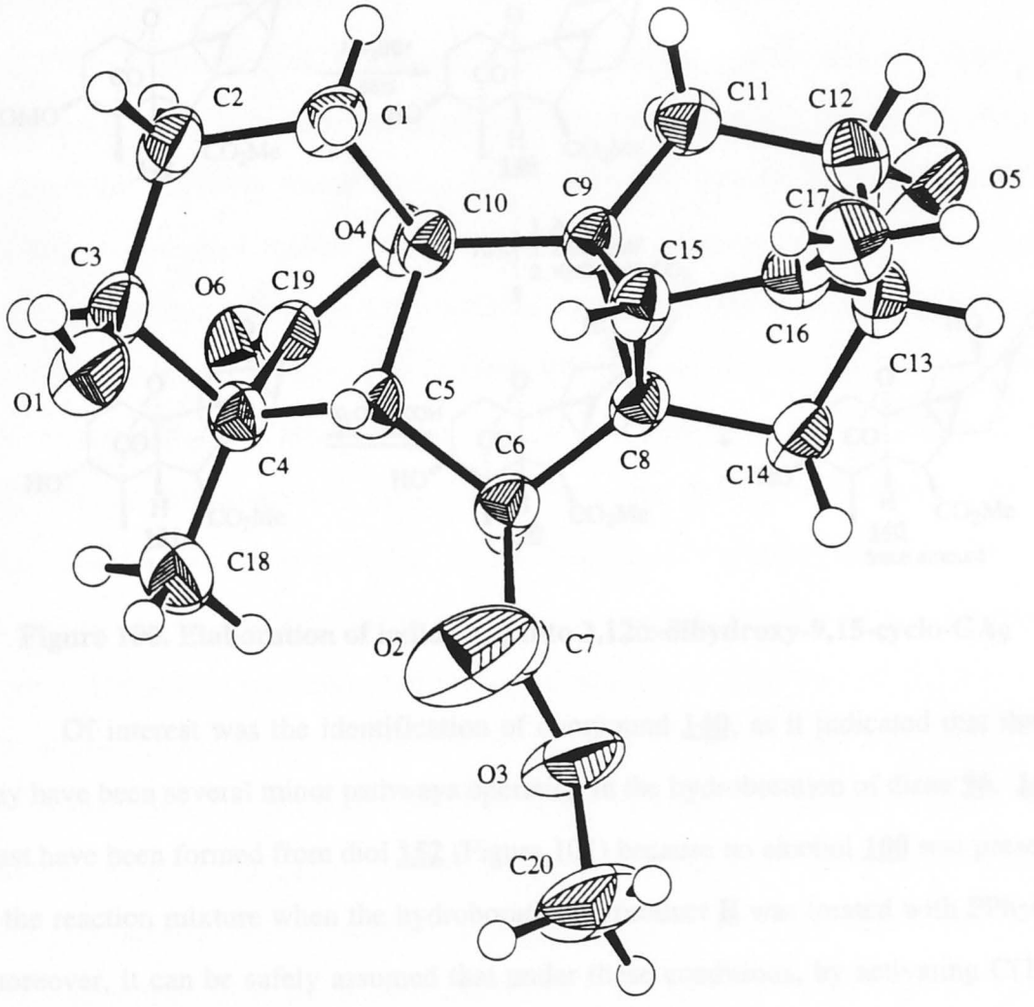
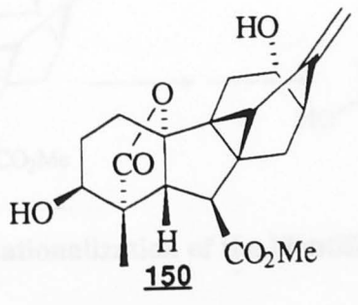


Figure 98. Reaction of mixture B with PPh_3/I_2

The 12-hydroxy derivative **148** turned out to be stereochemically homogeneous with only a trace amount of a contaminant inseparable by chromatography being observed in the ^{1}H NMR spectrum. The chemical shifts and the coupling pattern of the 17-methylene protons were indicative of the absolute configuration at C(16) being *S* (analogous to diol **100**). The original suspicion that the 12-hydroxy group was in the α configuration was given further support upon locating the doublet of H(14α) in the spectrum. This signal was shifted significantly downfield relative to compounds in the 12β-hydroxy series, a difference that was easily explained in terms of deshielding of H(14α) by the 12α-hydroxy group. The structure of **148** led to the assignment of the stereochemistry of the parent diol **147**.

With compound **148** in hand, the 12α-series could be prepared as well thereby completing the studies on 3,12-dihydroxy-9,15-cyclo-GA$_9$. Compound **148** was treated[86] with Me_2BBr and the diol **149** was converted into the olefin **150** using the same procedures as in the case of its diastereomer **139**. The ease with which **149** (protected as the diacetate) underwent the DBU-induced elimination was another indirect proof that the configuration at C(16) was *S*. After 5 hours, complete conversion was achieved. Base-catalysed deprotection afforded the desired target **150** together with a tiny amount of the 12β-hydroxy compound **140** which thus turned out to be the contaminant observed in the ^{1}H NMR spectrum of **148** (*vide supra*). Alcohol **150** was crystallised and its structure confirmed by single crystal X-ray analysis (Figure 99). As with **140**, compound **150** was partially epimerised[89] at C(3) in order to obtain a sufficient amount of the 3α,12α-dihydroxy isomer for GC-MS analysis (Figure 100).





150

Figure 99. Crystal structure of compound 150

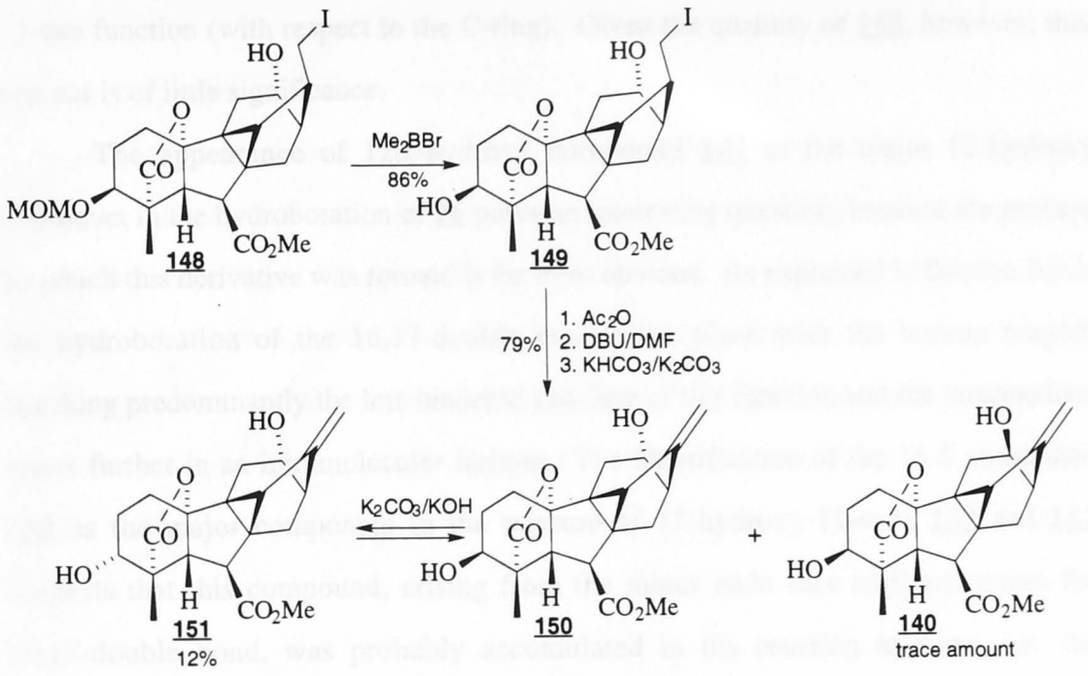


Figure 100. Elaboration of iodide 148 into 3,12α-dihydroxy-9,15-cyclo-GA$_9$

Of interest was the identification of compound **140**, as it indicated that there may have been several minor pathways operating in the hydroboration of diene **96**. **140** must have been formed from diol **152** (Figure 101) because no alcohol **100** was present in the reaction mixture when the hydroboration byproduct **B** was treated with PPh_3/I_2 (moreover, it can be safely assumed that under these conditions, by activating C(17) towards nucleophilic attack, any **100** would have been converted into the cyclic ether **113**, as outlined in Figure 75, Section 3.3.3).

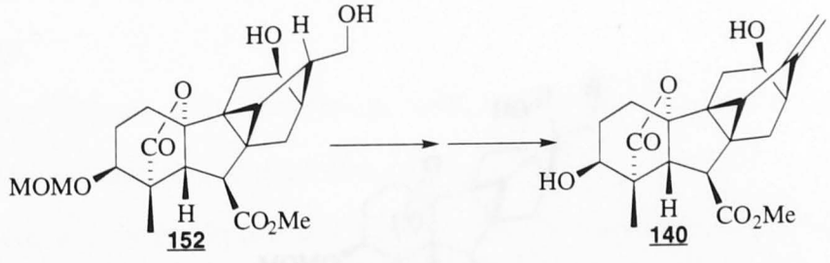
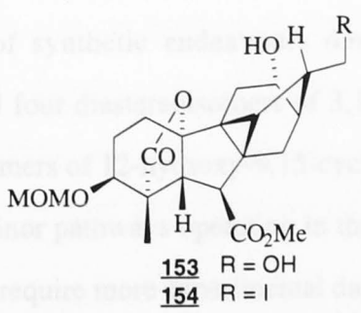


Figure 101. Rationalization of the identification of 140

The pathway for the formation of **152** is apparent: the 16,17-double bond in diene **96** was hydroborated on the more hindered endo-face and this process was followed by the addition of another molecule of the reagent across the top face of the

11-ene function (with respect to the C-ring). Given the quantity of **140**, however, this process is of little significance.

The appearance of 12α-hydroxy compound **147** as the major 12-hydroxy byproduct in the hydroboration of **96** poses an interesting question, because the process by which this derivative was formed is far from obvious. As explained in Section 3.3.1, the hydroboration of the 16,17-double bond takes place with the borane reagent attacking predominantly the less hindered exo-face of this function and the intermediate reacts further in an intramolecular fashion. The identification of the 16 *S* compound **142** as the major component in the mixture of 17-hydroxy-11-enes **142** and **143** suggests that this compound, arising from the minor endo face addition across the 16,17-double bond, was probably accumulated in the reaction mixture, *i.e.* the remaining 11-ene function did not react with the borane complex to an appreciable extent. The results obtained upon attempted reaction of **118** (Section 3.3.4.3) with $BH_3.SMe_2$ support the conclusion that the 11,12-double bond is relatively unreactive, unless the reagent is delivered intramolecularly. Besides, it was shown that in this reaction, the minor 16 *S* epimer corresponding to **142** was more reactive under forcing conditions than its 16 *R* counterpart, analogous to **143**. One would thus expect that if **147** was formed from the borane intermediate to **143** by an intermolecular α-face hydroboration followed by oxidation, so would have been its 16-epimeric counterpart **153** (Figure 102), which would have given rise to iodide **154**.



153 R = OH
154 R = I

Figure 102. Structures of hypothetical diol 153 and iodide 154

However, no evidence for the presence of this compound was observed in the spectra of iodide **148**, which was the only significant 12-hydroxy compound formed

upon treatment of byproduct **B** with PPh_3/I_2. A question therefore arises whether in fact the 12,17-diol **147** was not formed by the addition of $BH_3.SMe_2$ to the α-face of the 11-ene function in diene **96** first, which may have been made possible by the more favourable geometry of the 11,16-diene arrangement allowing a slightly better access to the 11,12-double bond. This process would be followed by the second addition to the 16-ene function, the exo-selectivity of which would be consistent with obtaining diol **147** from the fraction of diene molecules which was attacked from the bottom face of the C-ring (Figure 103). The resulting bis(borane) intermediate **156** would afford alcohol **147** upon oxidative work-up.

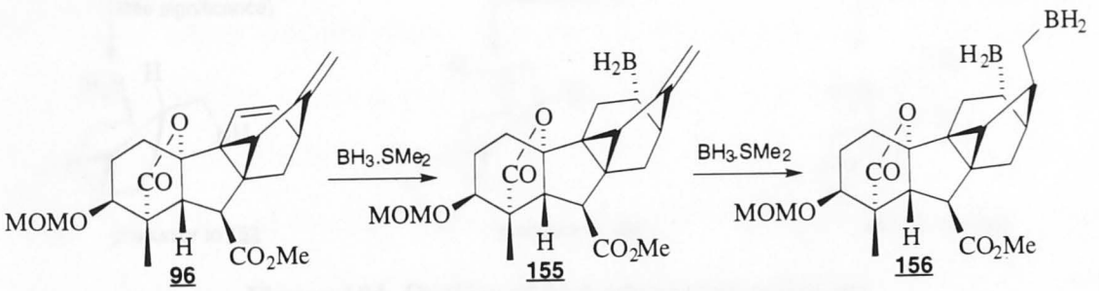


Figure 103. Possible hydroboration pathway leading to diol 147

In this regard, the major hydroboration route on which this approach is based may not have been the exclusive way leading to the major product of this reaction **100**: initial addition across the β-face of the 11-ene function and subsequent exo-hydroboration of the 16,17-double bond would have given **100** as well. All hydroboration pathways leading to 12-hydroxy compounds are summarised in Figure 104.

The primary goal of synthetic endeavours described in this Chapter was achieved by synthesizing all four diastereoisomers of 3,12-dihydroxy-9,15-cyclo-GA_9 methyl ester and the two epimers of 12-hydroxy-9,15-cyclo-GA_9 methyl ester. A more detailed discussion of the minor pathways operating in the hydroborations of dienes **96** and **103** would undoubtedly require more experimental data to be obtained.

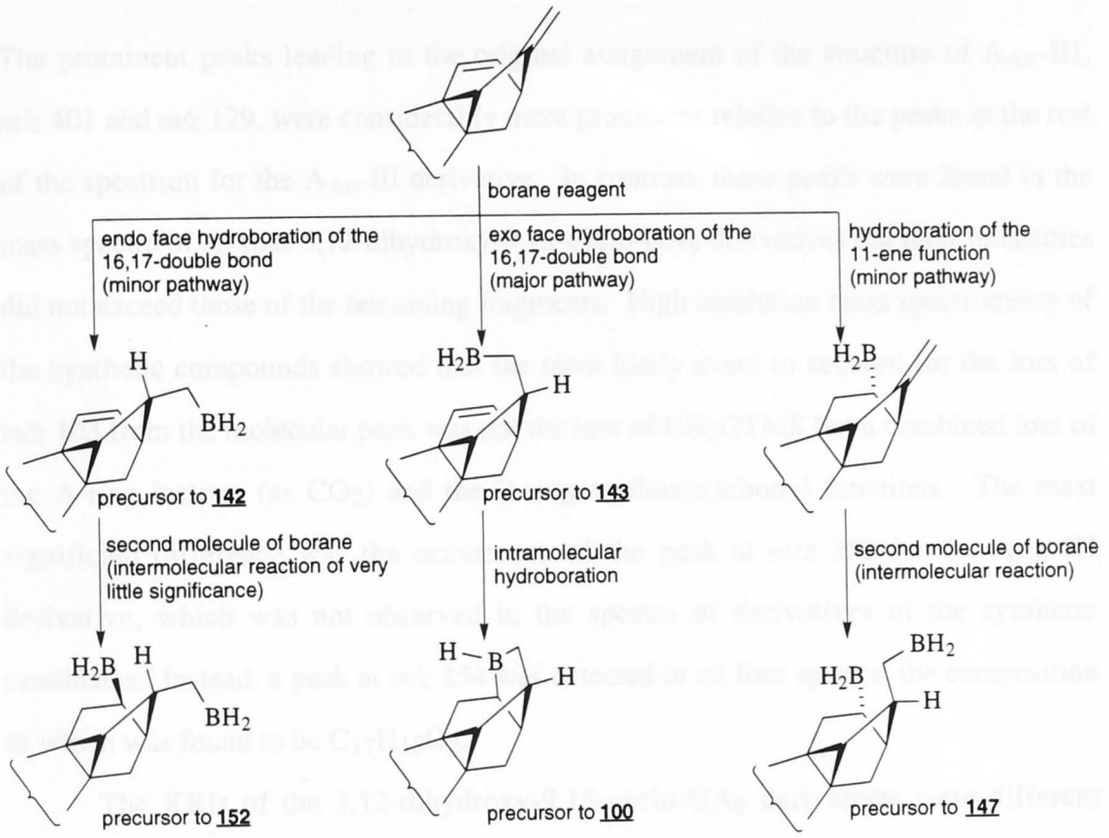


Figure 104. Outline of hydroboration pathways

3.3.7 GC-MS analysis

GC-MS comparisons of 3-deoxy compounds **135** and **137** to the metabolites isolated during the biosynthetic experiments on the 9,15-cyclo-GA_9 precursor **7** identified MI and LMII as 12α-hydroxy-9,15-cyclo-GA_9. Another metabolite from *Lygodium japonicum,* which was not referred to in Chapter 1 because it had been formed in a minor amount, was found to be 12β-hydroxy-9,15-cyclo-GA_9. These results confirmed the original suspicion that the C-ring position which was hydroxylated by *A. Phyllitidis* and *L. japonicum* enzymes in [$^2H_2$] 9,15-cyclo-GA_9 **7** was C(12).

It was therefore disappointing to find that A_{An}-III could not be assigned as one of the four diastereoisomers of 3,12-dihydroxy-9,15-cyclo-GA_9 beyond doubt. The mass spectra of the 3,12-dihydroxy-9,15-cyclo-GA_9 derivatives displayed a pattern similar to the A_{An}-III derivative. A few discrepancies, however, could be observed.

The prominent peaks leading to the original assignment of the structure of A_{An}-III, *m/z* 401 and *m/z* 129, were considerably more prominent relative to the peaks in the rest of the spectrum for the A_{An}-III derivative. In contrast, these peaks were found in the mass spectra of all four 3,12-dihydroxy-9,15-cyclo-GA_9 derivatives but their intensities did not exceed those of the remaining fragments. High resolution mass spectrometry of the synthetic compounds showed that the most likely event to account for the loss of *m/z* 103 from the molecular peak was not the loss of CH_2OTMS but a combined loss of the A-ring lactone (as CO_2) and the B-ring methoxycarbonyl functions. The most significant difference was the occurrence of the peak at *m/z* 252 for the A_{An}-III derivative, which was not observed in the spectra of derivatives of the synthetic candidates. Instead, a peak at *m/z* 254 was detected in all four spectra, the composition of which was found to be $C_{17}H_{18}O_2$.

The KRIs of the 3,12-dihydroxy-9,15-cyclo-GA_9 derivatives were different from that of the A_{An}-III derivative. 3α,12β-bis(trimethylsilyloxy)-9,15-cyclo-GA_9 methyl ester (2777) was the closest to the A_{An}-III derivative (2795).

These results put into question the correctness of the structural assignment of the basic skeleton of A_{An}-III and, consequently, the location of at least one of the hydroxy groups, if not both. Despite a number of similarities in the mass spectra of the four derivatives of the synthetic compounds and the A_{An}-III derivative, it did not appear possible to postulate an alternative structure for A_{An}-III.

In view of these findings, hydroxylation of the 9,15-cyclo-GA_9 precursor **7** by *A. phyllitidis* and *L. japonicum* enzymes may be ascribed to non-specific metabolism due to a high dose of substrate. This conclusion is also supported by the fact that, apart from antheridic acid, no dihydroxy compounds which would correspond to A_{An}-III were identified in the culture medium from the metabolic studies on 9,15-cyclo-GA_9.

CHAPTER 4

4. SYNTHESIS OF 12-HYDROXY-GA$_{73}$ METHYL ESTER

4.1 INTRODUCTION

While the studies on 12-hydroxy-9,15-cyclo-GA$_9$ described in the previous Chapter were in progress, Yamane and his co-workers[90] isolated two very similar neutral antheridiogens from the prothallia of the fern, *Lygodium circinnatum*. GC-MS analysis suggested that they were of the GA$_{73}$ methyl ester type (**4**) with an additional hydroxy group. As the most likely location of the hydroxy group was C(12), the two compounds were tentatively assigned as 12α- and 12β-hydroxy-GA$_{73}$ methyl ester **157** and **158** , respectively (Figure 105).

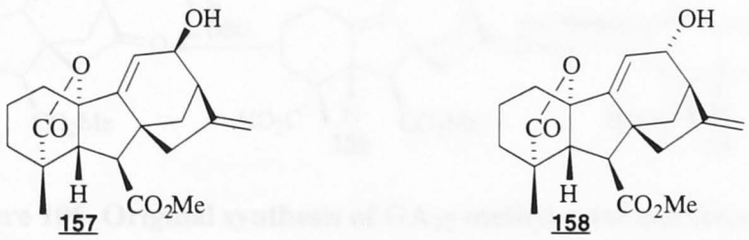


Figure 105. Tentative assignment of the structures of the new antheridiogens

It is more than obvious that **157** and **158** are structurally related to 12-hydroxy-9,15-cyclo-GA$_9$ methyl ester epimers **135** and **137**, sharing the same hydroxylation pattern with the 9,11-double bond being isomeric to the 9,15-cyclo function. Structure elucidation of the two new antheridiogens by partial synthesis is thus closely linked to the studies described in this thesis.

From a synthetic point of view, the preparation of **157** and **158** did not seem to pose a major challenge, as it appeared that these compounds could be conveniently obtained by an allylic functionalization of a suitable 9,(11)-ene precursor. This consideration, in fact, dictated the pathway to be explored first: an allylic functionalisation of GA$_{73}$ methyl ester norketone **160** followed by elaboration towards the target compounds *via* functional group interconversions (*vide infra*).

As already indicated in Chapter 1, GA_{73} methyl ester norketone was an advanced intermediate in the synthesis by Kraft-Klaunzer and Mander of GA_{73} methyl ester (**4**) from GA_7 methyl ester[36]. This sequence was based on the extension of functionality from the A-ring into the C-ring, but in view of findings made during studies on 11-hydroxy and 12-hydroxy-9,15-cyclo-GA_9, it could be considerably improved (Figure 106).

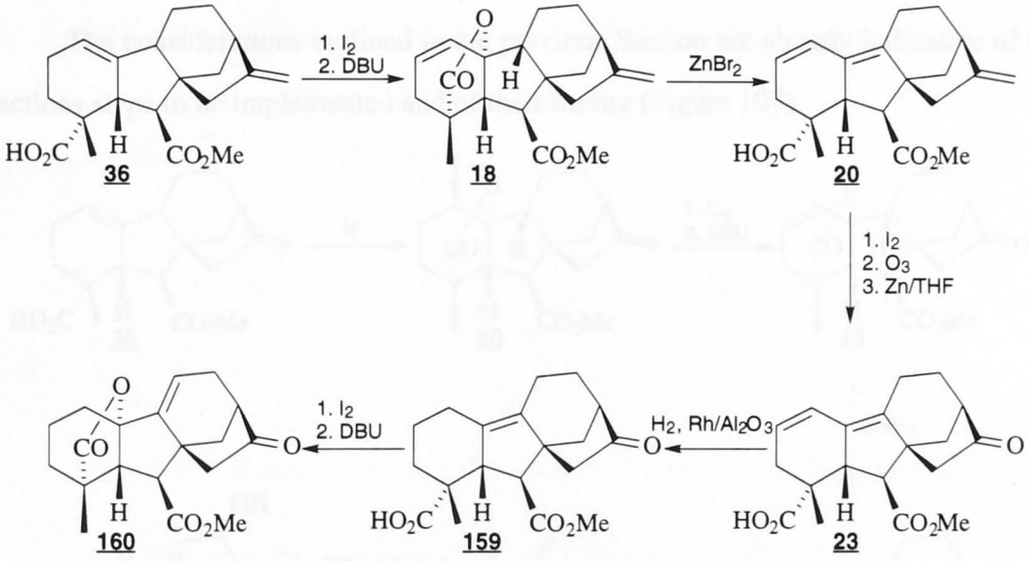


Figure 106. Original synthesis of GA_{73} methyl ester norketone 160

An apparent drawback of the original synthesis is the iodolactonization/ ozonolysis/ reductive elimination cycle (**20** → **23**), which was employed to allow selective reduction of the 1,2-double bond, thus adding a few extra steps to the sequence. The more direct approach to **23**, based on the preparation of the 16-oxo analogue of compound **18**, followed by treatment of this derivative (**21**, Section 1.2.3, Chapter 1) with $ZnBr_2$, was also attempted[36,37]. This route was abandoned at that time, however, due to the formation of the equilibrium mixture of the parent compound **21**, the isomeric lactone **22** and the desired product **23** upon treatment of **21** with $ZnBr_2$ in ether, as described in Chapter 1.

Clearly, the protocol developed for the rearrangement of **81** into the diene acid **82** (see Section 3.2.2, Chapter 3) with moist $ZnBr_2$ provided the means for resolving this problem. Similarly, the early intermediate **36** could be prepared from the more readily accessible GA_3 methyl ester *via* the dimesylate **48**.

A minor project was therefore mounted with a view to improving the efficiency of the original preparation of GA_{73} methyl ester norketone and broadening this sequence by synthesizing 12-hydroxy-GA_{73} methyl ester.

4.2 SYNTHETIC PLAN

The considerations outlined in the previous Section are already indicative of the reactions steps to be implemented and of their timing (Figure 107).

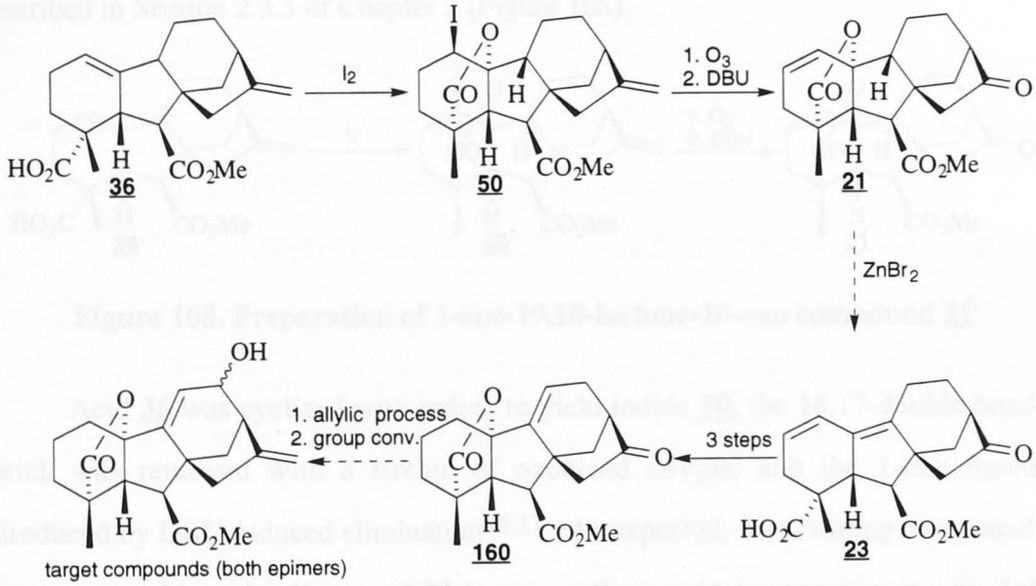


Figure 107. Synthetic plan

The starting material, acid **36** could be converted[36,37] into the 16-oxo analogue of compound **18** (**21**, cf. Chapter 2, Section 2.3.3). This 1-ene-16-oxo compound was expected to undergo a complete rearrangement into the diene acid **23** upon treatment with moist $ZnBr_2$, as described for the conversion of **81** into **82**. The desired GA_{73} methyl ester norketone could be obtained from **23** in a straightforward manner over three steps involving catalytic hydrogenation, iodolactonisation and elimination[36]. Subsequent allylic functionalisation of **160** followed by functional group interconversions would produce, at the final stage, the target compound(s).

The synthesis can therefore be classified as a type 1 process (Section 1.2.2, Chapter 1); the 9,(11)-ene function introduced by the transposition of the A-ring functionality is extended to a further position in the C-ring.

4.3 IMPLEMENTATION OF THE PLAN

The starting material, acid **36**, was prepared from the dimesylate **48** (Chapter 2, Section 2.3.2) and elaborated towards intermediate **21** according to procedures described in Section 2.3.3 of Chapter 2 (Figure 108).

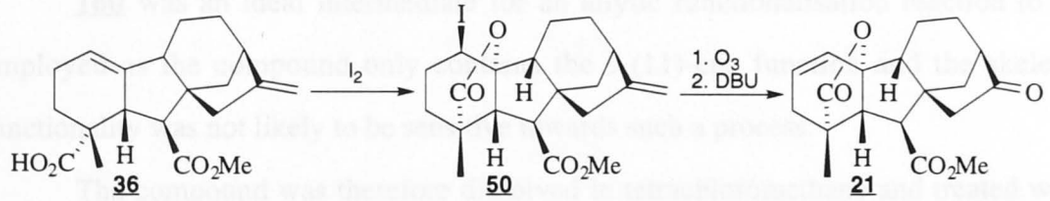


Figure 108. Preparation of 1-ene-19,10-lactone-16-oxo compound 21

Acid **36** was cyclized with iodine to yield iodide **50**, the 16,17-double bond of which was removed with a stream of ozonised oxygen and the 1-ene function introduced by DBU-induced elimination[36,37]. As expected, the resulting compound **21** was rearranged into the diene acid **23** in an excellent yield upon treatment with $ZnBr_2$ in ether when the reaction mixture was allowed to gradually absorb atmospheric moisture. TLC monitoring of the process revealed that the completion of the reaction coincided with the complete dissolution of $ZnBr_2$ whereby the reaction mixture became homogeneous (Figure 109).

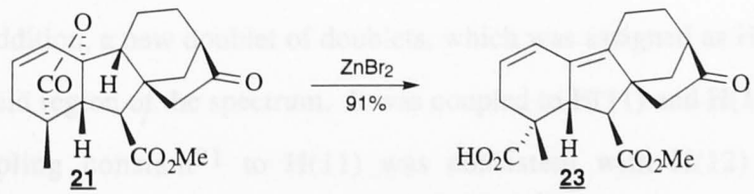


Figure 109. $ZnBr_2$-mediated rearrangement of 21

Having resolved the problem of the $ZnBr_2$-induced rearrangement of compound **21**, GA_{73} methyl ester norketone **160** could be synthesized by a three-step sequence:

selective hydrogenation of the 1,2-double bond, followed by the iodolactonisation of the carboxy group onto the 9-ene function and DBU elimination of the elements of HI from the C-ring[36] (Figure 110). **160** was thus prepared in 7 steps from **36** as compared to the original synthesis, which required 9 steps to achieve the same overall conversion.

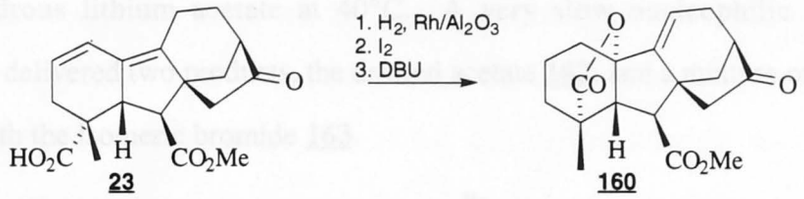


Figure 110. Completion of the shortened synthesis of 160

160 was an ideal intermediate for an allylic functionalisation reaction to be employed as the compound only contains the 9,(11)-ene function and the skeletal functionality was not likely to be sensitive towards such a process.

The compound was therefore dissolved in tetrachloromethane and treated with N-bromosuccinimide and a catalytic amount of dibenzoyl peroxide as the initiator under reflux. The reaction afforded a single product, the ^1H NMR spectrum of which corresponded to structure **161** (Figure 111).

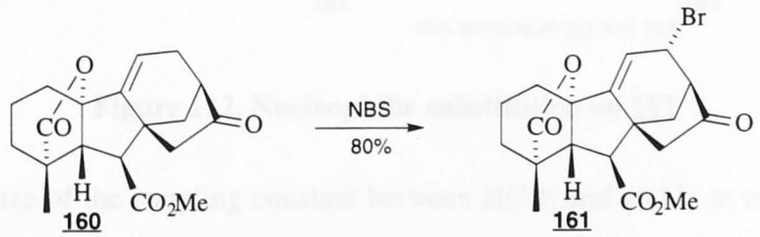


Figure 111. Allylic bromination of 160

The signal of H(11) shifted from 5.86 ppm in the parent compound to 6.10 ppm in **161**. In addition, a new doublet of doublets, which was assigned as H(12), appeared in the low field region of the spectrum. It was coupled to H(11) and H(13) and the size of the coupling constant[91] to H(11) was consistent with H(12) being in the β-configuration. The ^{13}C spectrum, mass spectrum (including HRMS) and elemental analysis were fully in accord with this assignment. The α-face selectivity of the bromination of the initially formed allylic radical contrasts with a similar allylic bromination (**15** → **17**) described in Chapters 1 and 2 and is probably of steric origin.

With the new synthetic function in place, the target compounds could be prepared by the conversion of the Br(12α)-substituent into the hydroxy function and the 16-keto group into the 16-ene moiety. The obvious method to achieve the former transformation was an S_N2 process and **161** was thus dissolved in dry DMF and treated with anhydrous lithium acetate at 40°C. A very slow nucleophilic substitution (*ca* 3 days) delivered two products, the desired acetate **162**, and a mixture of the starting material with the isomeric bromide **163**.

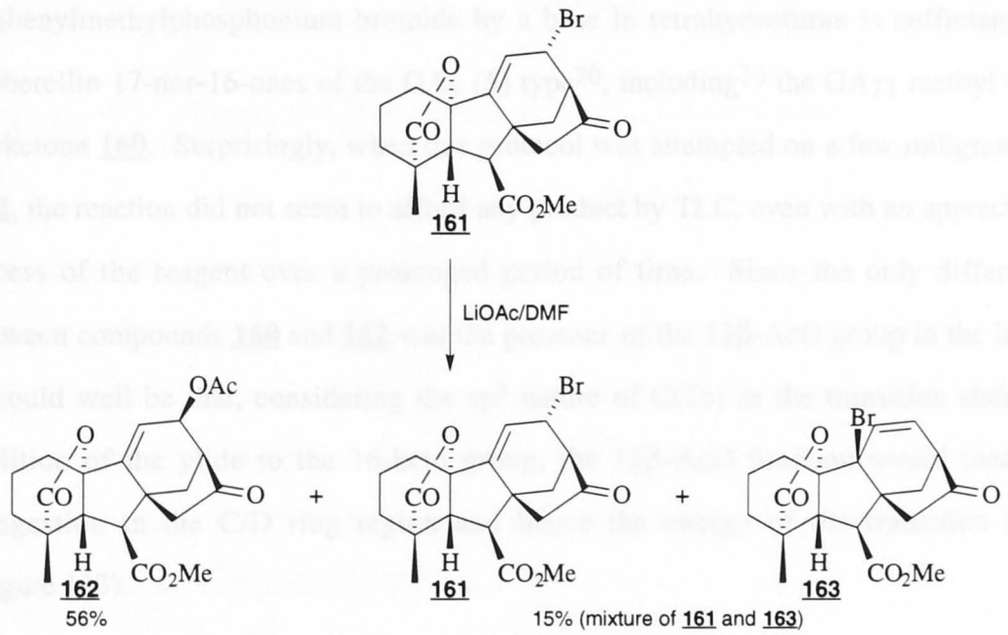


Figure 112. Nucleophilic substitution on 161

The size of the coupling constant between H(12) and H(11) in compound **162** (2.9 Hz) indicated the α-configuration of H(12), as estimated from the inspection of a Dreiding model.

The ^1H NMR spectrum of bromide **163** which was formed in an S_N2'-like process displayed two mutually coupled olefinic signals of H(11) (6.28 ppm, doublet) and H(12) (6.03 ppm, triplet). The diagnostic resonance of H(5) was found at 2.94 ppm (Δ 0.5 ppm downfield relative to H(5) in **161** and therefore consistent with a 9β-bromo substituent), while the doublet of H(6) was shifted to higher field relative to its counterpart in the parent compound **161**. The identification of this substance in the reaction mixture suggests that C(12) is not easily approachable by the nucleophile. Apparently, the allylic cation, arising from the dissociation of Br^-, and the leaving Br^-

must exist as an ionic pair for a considerable amount of time which allows the bromine nucleophile to attack C(9). This is in accord with the sterically congested nature of the C/D ring region and the slow rate of the overall process.

The major product **162** could be subsequently subjected to a suitable methylenation procedure followed by the hydrolysis of the 12-acetoxy function to obtain the target molecules. It had been well established that a simple version of the Wittig methylenation reaction based on the generation of the ylide from triphenylmethylphosphonium bromide by a base in tetrahydrofuran is sufficient for gibberellin 17-nor-16-ones of the GA_9 (**5**) type[20], including[36] the GA_{73} methyl ester norketone **160**. Surprisingly, when this protocol was attempted on a few miligrams of **162**, the reaction did not seem to afford any product by TLC, even with an appreciable excess of the reagent over a prolonged period of time. Since the only difference between compounds **160** and **162** was the presence of the 12β-AcO group in the latter, it could well be that, considering the sp^3 nature of C(16) in the transition state for addition of the ylide to the 16-keto group, the 12β-AcO function would increase congestion in the C/D ring region and hence the energy of the transition state (Figure 113).

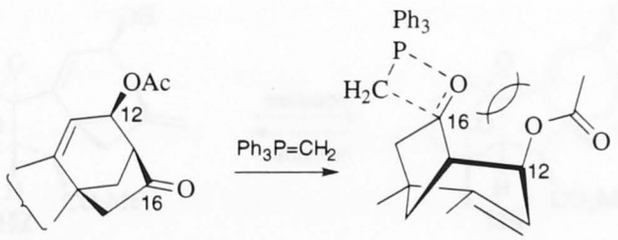


Figure 113. Possible steric interactions in the transition state

Given the amount of **162** which was available, it was decided to employ the Lombardo-Oshima methylenation procedure[63] instead of optimizing the conditions of a Wittig-type process. Although it could not be expected that this reaction would provide a good yield, in view of the presence of the sensitive allylic A-ring lactone function, it was almost certain to effect some of the desired conversion.

In the event, the $Zn/CH_2Br_2/TiCl_4$ methylenation restored the 16-ene function and the resulting derivative **164** was deprotected with base to afford one of the target

molecules **157**, the yield of the methylenation being 20%. The structures of byproducts in the methylenation step were not analyzed as they formed a complex mixture of polar compounds.

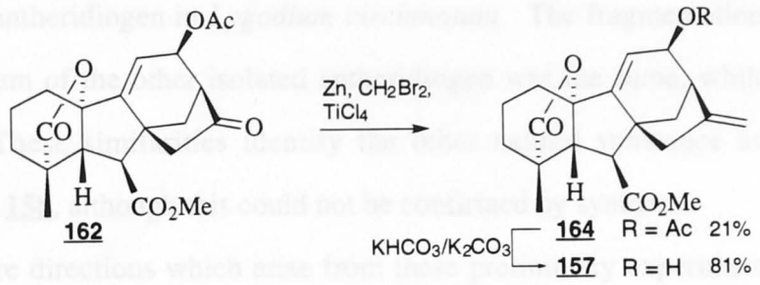


Figure 114. Restoration of the 16,17-double bond

At this juncture, the amount of material for further work was considerably reduced (*ca* 4 mg) and only high yielding reactions could be attempted to address the problem of inverting the configuration at C(12), an oxidation-reduction cycle being a logical choice. Alcohol **157** was therefore oxidized into ketone **165** with the Dess-Martin periodinane[64], but the reduction of **165** with $NaBH_4$ proceeded with 100% α-face selectivity, thereby returning the β-alcohol, regardless of the reaction conditions ($NaBH_4$/MeOH; $NaBH_4$, $CeCl_3$/MeOH).

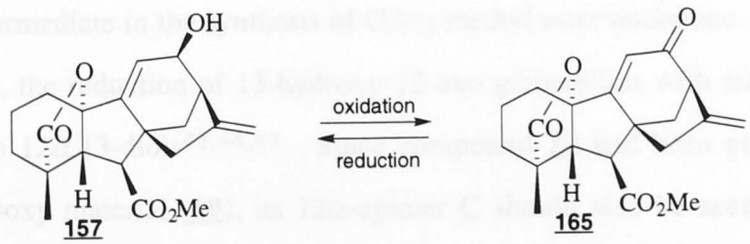


Figure 115. Oxidation/reduction cycle on alcohol 157

Not even a trace of the epimeric 12α-alcohol was observed in the ^1H NMR spectrum. This outcome is not surprising as it falls in line with the results obtained upon reductions of 12-ketones in gibberellin and 9,15-cyclogibberellin compounds described in Chapter 3.

4.4 GC-MS ANALYSIS

GC-MS comparison of the derived trimethylsilyl ethers showed that alcohol **157** is a natural antheridiogen in *Lygodium circinnatum*. The fragmentation pattern in the mass spectrum of the other isolated antheridiogen was the same, while the KRI was different. These similarities identify the other natural substance as the epimeric 12α-alcohol **158**, although this could not be confirmed by synthesis.

Future directions which arise from these preliminary experiments are twofold. The conversion of the 16-oxo function in the norketone **162** into the corresponding double bond could be carried out more efficiently as long as suitable conditions for a Wittig-type process are found and optimized. Secondly, due to the failure of the reduction of ketone **165** to deliver the 12α-alcohol **158**, a suitable synthesis of this compound should be developed.

It appears that a synthetic route to **158** could be opened up based on the results obtained during the initial studies towards 12-hydroxy-9,15-cyclogibberellins. 12β-hydroxy derivative **74** which was converted into the diene acid **82** (Section 3.2.2, Chapter 3) over a few steps in this synthesis is a 12-hydroxy analogue of compound **36**, the early intermediate in the synthesis of GA_{73} methyl ester norketone. As mentioned in Chapter 3, the reduction of 13-hydroxy-12-oxo gibberellins with zinc borohydride gives rise to 12α,13-diols[43,44,45]. Since compound **74** had been prepared from a 12,13-dihydroxy material (**69**), its 12α-epimer **C** should also be accessible and the design of the synthetic pathway is obvious:

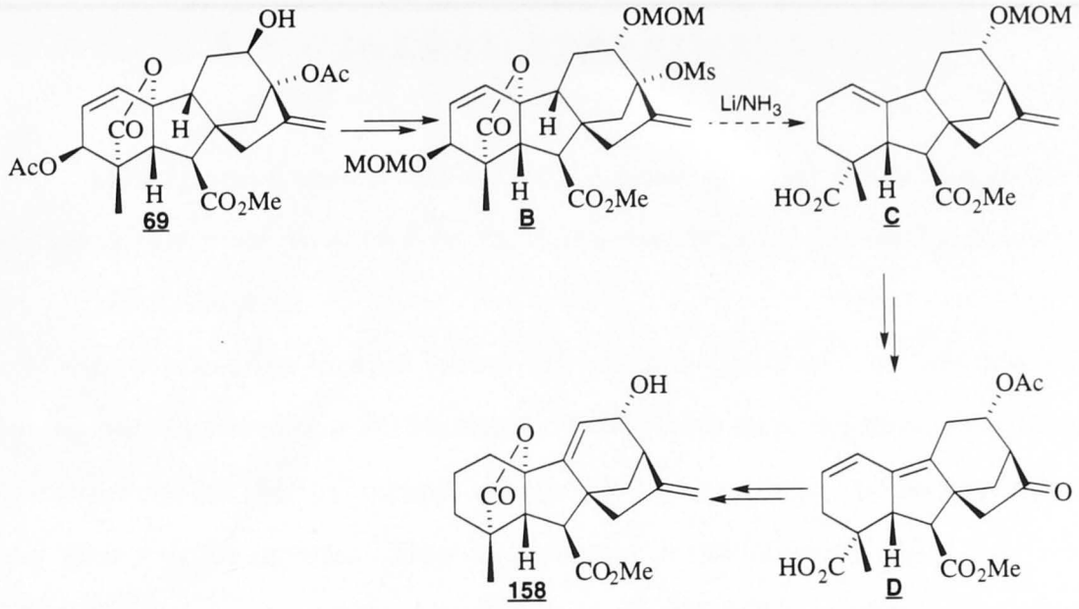


Figure 116. Designed synthetic pathway to <u>158</u>

The first part of the synthesis, utilizing compound **<u>69</u>** as the starting material, involves the inversion of configuration at C(12) combined with functional group manipulation in order to achieve a suitable arrangement (**<u>B</u>**) for a Birch-type deoxygenation of C(13) and C(3) (Section 2.3.2 of Chapter 2). This sequence would produce derivative **<u>C</u>**, epimeric with **<u>74</u>**. The second part (**<u>C</u>** → **<u>D</u>**) can be executed in a fashion analogous to the synthesis of the diene acid **<u>82</u>** (Section 3.2.2 of Chapter 3) and the sequence could be completed by the conversion of **<u>C</u>** into the target compound **<u>158</u>** as in the synthesis of GA_{73} methyl ester[36].

EXPERIMENTAL

5.1 GENERAL EXPERIMENTAL

Melting points were determined on a Reichert hot-stage and are uncorrected. Microanalyses were performed by the Australian National University Analytical Services Unit, Canberra.

Low resolution EI mass spectra (70 eV) and high resolution accurate mass spectra were measured on a VG Micromass 7070F double focussing mass spectrometer. The molecular ion ($M^{\cdot+}$), if present, significantly high mass ions and the more intense low mass ions are reported. Data are presented in the following order: *m/z* value; relative intensity as a percentage of the base peak. GC-MS comparisons of synthetic samples with natural substances were carried out by Associate Professor H. Yamane at the Biotechnology Research Centre, University of Tokyo. Prior to being subjected to GC-MS analysis, the synthetic alcohols were converted into the trimethylsilyl derivatives by treatment with BSTFA/1% TMSCl in pyridine.

Infrared spectra (ν_{max}) were recorded in $CDCl_3$ on a Perkin-Elmer 683 Infrared spectrophotometer in 0.25 mm NaCl solution cells.

The optical rotations ($[\alpha]_D^{20}$) were measured in CH_2Cl_2 (unless otherwise stated) with a Perkin-Elmer 341 polarimeter at 20°C with an error of $\leq \pm 1°$.

The NMR spectra were recorded for $CDCl_3$ solutions at 25°C on the following instruments: Varian Gemini 300 (operating at 300 MHz for ^1H, 75.5 MHz for ^{13}C), Varian VXR 300 (operating at 300 MHz for ^1H, 75.4 MHz for ^{13}C) and Varian VXR 500 (operating at 500 MHz for ^1H). Chemical shifts were recorded as δ values in parts per million (ppm) and were indirectly referenced to tetramethylsilane (TMS) *via* the solvent signals (7.26 ppm for ^1H and 77.0 ppm for ^{13}C). Data were recorded as follows: chemical shift (δ), integrated intensity (for proton spectra), multiplicity (**s**: singlet, **d**: doublet, **t**: triplet, **q**: quartet, **m**: multiplet, **dd**: doublet of doublets, etc: **br** indicates some degree of broadening in the signal), coupling constant(s) (Hz), and assignment (first order analyses of spectra were attempted where possible and, consequently, chemical shifts and coupling constants for multiplets may only be approximate). Where ^1H NMR spectra of mixtures were recorded, only those data

which could be determined unequivocally were given. Attached proton test (APT) experiment[76] was used in the assignment of carbon atoms. When an unequivocal assignment could not be made, the multiplicity of a carbon was indicated by the number of attached protons. In the cases where CH_3 and CH or CH_2 and C atoms could not be discriminated, the multiplicity was described as either even (e) or odd (o). Distortionless enhancement by polarization transfer[76] (DEPT) and two-dimensional NMR experiments (*vide infra*) were used to assign the molecular framework of compound **100** and related derivatives.

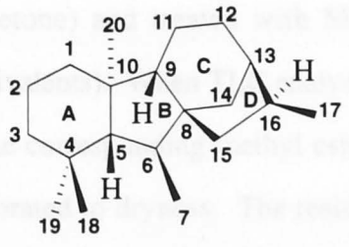
Two dimensional NMR experiments[76] were carried out using the following instruments: Varian VXR 300 and Varian VXR 500. The pulse sequences used were double quantum filtered homonuclear ($^1H/^1H$) correlation spectroscopy (DQF COSY), heteronuclear ($^1H/^{13}C$) correlation spectroscopy (HETCOR), long range heteronuclear ($^1H/^{13}C$) correlation spectroscopy (LR HETCOR), total correlation spectroscopy (TOCSY) and the combination of heteronuclear multiple quantum coherence spectroscopy (HMQC) and TOCSY sequences. With the exception of the HMQC TOCSY experiment, standard software supplied by the manufacturer was used throughout. A default value of 8 Hz for $^nJ_{CH}$ (n = 2, 3, ...) was used in the LR HETCOR sequence (compound **100**). The mixing time in the TOCSY sequence was 40 ms (compound **100**). The HMQC TOCSY spectrum acquired utilized the pulse sequence of Lerner and Bax[77], modified as described in the review of Martin and Crouch[78]; the mixing time was 30 ms (compound **100**).

Analytical thin layer chromatography (TLC) was conducted on micro-slides coated with Merck Kieselgel KG60F-254. The developed plates were visualised under short-wave ultraviolet light and stained with 13% (w/v) vanillin in concentrated sulfuric acid at 180°C. Flash chromatography was conducted according to the method of Still[93] *et al.* using Merck Kieselgel 60 as the adsorbent and analytical reagent (AR) grade solvents as indicated.

Solvents and reagents used in the reactions were purified according to well-established procedures[94]. Tetrahydrofuran (THF), diethyl ether (ether) and benzene were purified by distillation from sodium benzophenone ketyl.

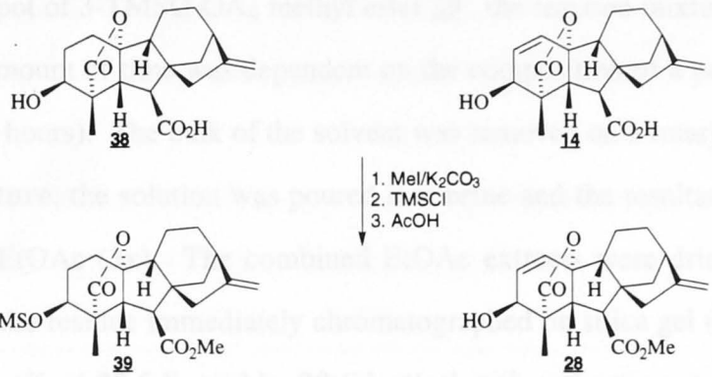
N,*N*-Dimethylformamide (DMF) was dried by the method of Burfield and Smithers[95]. Dichloromethane and triethylamine were distilled from calcium hydride. Unless otherwise stated, all reactions requiring dry solvents were performed under a dry nitrogen atmosphere. All reactions were carried out at ambient temperature, unless otherwise noted. Reaction temperatures refer to the external bath temperature. Standard work-up of an ethyl acetate or ethereal solution means washing with 1M HCl (aqueous), water and 5% $KHCO_3$ (aqueous). All organic extracts were dried with anhydrous sodium sulfate. After filtration of solutions from drying agent, the bulk of the solvent was removed on a Büchi rotatory evaporator and the last traces of solvent were removed under high vacuum. The identity of samples prepared by different routes was checked by TLC and IR and NMR spectra. Yields are given for isolated product showing one spot on a chromatographic plate and no impurities detectable in the NMR spectra.

All compounds referred to in the Experimental were named as the derivatives of *ent*-gibberellane:



5.2 CHAPTER 2 EXPERIMENTAL

General procedure for the derivatisation of the commercially available mixture of GA_7 and GA_4 and subsequent separation of GA_7 methyl ester 28



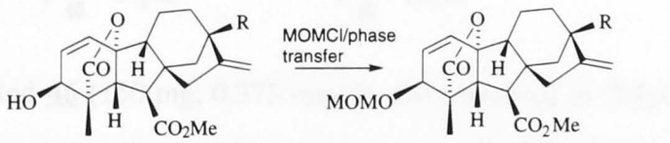
The commercially available GA_4/GA_7 mixture (as the molecular weights of the two compounds only differ by 2 molecular weight units, the mixture was, for the purpose of calculating the amounts of reagents, regarded as one component of the molecular mass of GA_7) was dissolved in acetone (1g of the mixture dissolves in approximately 20 ml of acetone) and treated with MeI (10-12 equivalents) in the presence of K_2CO_3 (2-4 equivalents). When TLC analysis showed that the highly polar acids were converted into the corresponding methyl esters (overnight), the slurry was filtered and the filtrate evaporated to dryness. The residue was redissolved in EtOAc, washed with brine, the organic phase dried and evaporated.

The solid material was redissolved in CH_2Cl_2 and the solution treated with TMSCl (4-6 equivalents) and Et_3N (5-7 equivalents). After the formation of the 3-TMSO derivatives reached completion, $Et_3NH^+Cl^-$ was removed by filtration and the solvent evaporated. The residue was redissolved in EtOAc and the solution washed successively with H_2O and brine. The solution was then dried and the solvent removed *in vacuo*.

The crude mixture of GA_4 and GA_7 derivatives was redissolved in MeOH (roughly 10 mg per 1 ml of the solvent) and the solution was cooled to 0°C. Glacial acetic acid (1 equivalent) was added and the mixture was stirred at this temperature with careful monitoring by TLC. Upon visualisation of the TLC plates with vanillin in

concentrated sulfuric acid followed by heating, the GA_4 derivative showed a red spot while the GA_7 compound appeared as a green spot. When the hydrolysis of the 3-TMSO-GA_7 derivative into GA_7 methyl ester **28** reached approximately 80-90% conversion, which was indicated by a tinge of green colour neighbouring on the lowest part of the red spot of 3-TMSO-GA_4 methyl ester **39** , the reaction mixture was worked up (the actual amount of time was dependent on the composition of a particular batch; generally 24-72 hours). The bulk of the solvent was removed on a rotary evaporator at **room temperature**, the solution was poured into brine and the resultant mixture was extracted with EtOAc (3x). The combined EtOAc extracts were dried, the solvent evaporated and the residue immediately chromatographed on silica gel (EtOAc/hexane 9:1 then 3:2) to afford **39** followed by **28** (identical with authentic samples[52]). These operations were routinely performed on a scale as large as 10 g. The purity of compound **28** was within the range of 86-92%, as determined by ^1H NMR and this material could be used without purification. The actual amount depended on the composition of a particular batch of GA_4/GA_7.

General procedure for the phase transfer reaction of 3-hydroxy gibberellins with MOMCl



28 R = H
45 R = OH

35 R = H 91-95%
46 R = OH 84-86%
47 R = OMOM 8-10%

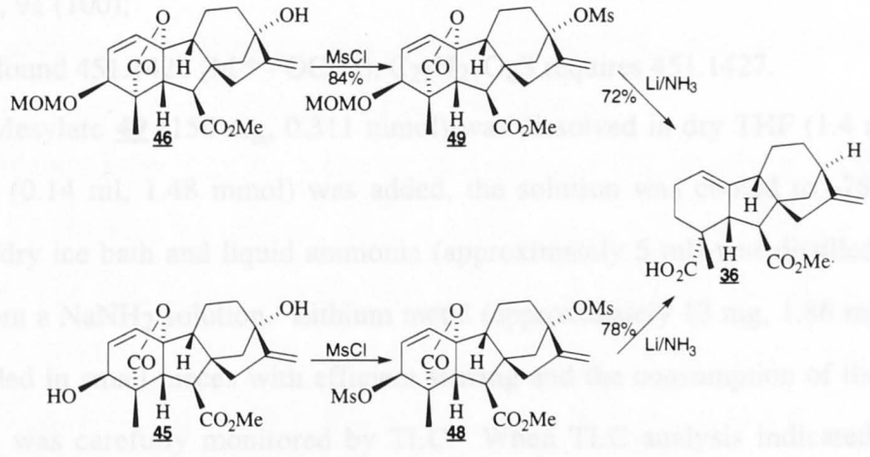
The 3-hydroxygibberellin derivative (7-methyl ester, 50-100 mg) was dissolved in benzene (4 ml, a mixture of benzene/EtOAc 3:1 was used for GA_3). $Bu_4N^+Br^-$ (1.2 equivalents) was added to the solution followed by 4M aqueous NaOH (8 ml) and the resulting two-phase mixture was cooled to 8-10°C. MOMCl (7-10 equivalents) was then added in one portion with efficient stirring. The conversion of the alcohol into the 3-methoxymethoxy derivative required 15-25 minutes. The mixture was transferred into a separating funnel and diluted with EtOAc and water. The inorganic phase was

extracted with EtOAc (3x), combined ethyl acetate extracts were concentrated under reduced pressure and the solution was filtered through a short column of alumina. The solvent was then removed and the residue chromatographed. This procedure appeared to be efficient on 10-100 mg scale. The attempts to scale it up resulted in obtaining appreciable amounts of byproducts.

GA_7 methyl ester: 3-MOMO-GA_7 methyl ester[36] (**35**, 91-95%), identical with an authentic sample;

GA_3 methyl ester: 3-MOMO-GA_3 methyl ester[96] (**46**, 84-86%) and 3,13-di-MOMO-GA_3 methyl ester[48] (**47**, 8-10%), identical with authentic samples.

Preparation of compounds 48 and 49 and the subsequent reaction with Li/NH_3



Compound **46** (150 mg, 0.373 mmol) was dissolved in CH_2Cl_2 (5 ml) and the solution was treated with Et_3N (0.5 ml, 3.59 mmol) and MsCl (0.15 ml, 1.94 mmol). A catalytic amount of DMAP was then added and the reaction mixture was allowed to stand for 16 hours. The solution was stirred with a few pieces of ice for 1 hour, the resulting mixture was diluted with EtOAc, transferred into a separating funnel and the organic layer separated. The solution was subjected to standard work-up, dried and the solvent removed (the last traces of the solvent and the reagent were removed under high vacuum at 50°C). Chromatography on silica gel (EtOAc/hexane 1:1) afforded the mesylate **49** (150 mg, 84%):

^{1}H NMR (300 MHz, $CDCl_3$) 6.28 (1H, d, $J_{1,2}$ = 9.4 Hz, H1), 5.95 (1H, dd, $J_{2,1}$ = 9.4 Hz, $J_{2,3\alpha}$ = 3.8 Hz, H2), 5.37 (1H, t, J = 2.4 Hz, H17), 5.13 (1H, s, H'17),

4.74 (1H, d, J = 6.9 Hz, -O$\underline{CH_2}OCH_3$), 4.67 (1H, d, J = 6.9 Hz, -O$\underline{CH_2}OCH_3$), 3.99 (1H, d, $J_{3\alpha,2}$ = 3.8 Hz, H3α), 3.75 (3H, s, -$CO_2\underline{CH_3}$), 3.38 (3H, s, -$OCH_2O\underline{CH_3}$), 3.31 (1H, d, $J_{5,6}$ = 10.7 Hz, H5), 3.03 (3H, s, -$OSO_2\underline{CH_3}$), 2.80 (1H, d, $J_{6,5}$ = 10.7 Hz, H6), 1.92 (1H, d, $J_{14\alpha,14\beta}$ = 11.8 Hz, H14α), 1.23 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 178.2 (CO), 172.3 (CO), 151.9 (C16), 132.3 (C2), 131.7 (C1), 110.7 (C17), 97.1 (C13), 91.6 (-O**C**H_2OCH_3 or C10), 90.3 (-O**C**H_2OCH_3 or C10), 75.1 (C3), 56.0 (-OCH_2O**C**H_3), 53.6 (CH), 53.4 (C), 52.4 (-CO_2**C**H_3), 51.3 (C), 50.6 (CH), 50.5 (CH), 42.4 (CH_2), 41.4 (CH_2), 41.1 (-OSO_2**C**H_3), 37.5 (CH_2), 17.1 (CH_2), 14.7 (C18);

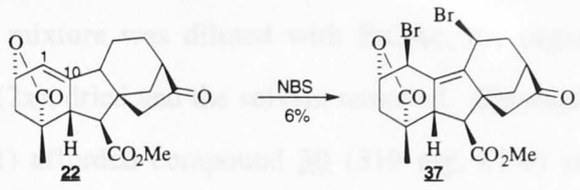
LRMS 451 ($M^{\cdot+}$ - OCH_3, 2), 420 (3), 376 (2), 354 (10), 324 (7), 316 (51), 297 (7), 280 (27), 265 (7), 237 (39), 221 (88), 209 (25), 193 (21), 179 (20), 165 (19), 155 (24), 129 (29), 91 (100);

HRMS found 451.1426 ($M^{\cdot+}$ - OCH_3), $C_{22}H_{27}O_8S$ requires 451.1427.

Mesylate **49** (150 mg, 0.311 mmol) was dissolved in dry THF (1.4 ml). Dry t-BuOH (0.14 ml, 1.48 mmol) was added, the solution was cooled to -78°C in an acetone/dry ice bath and liquid ammonia (approximately 5 ml) was distilled into the flask from a $NaNH_2$ solution. Lithium metal (approximately 13 mg, 1.86 mmol) was then added in small pieces with efficient stirring and the consumption of the starting material was carefully monitored by TLC. When TLC analysis indicated that the starting material had almost disappeared, the reaction was quenched with solid NH_4Cl and the ammonia allowed to evaporate. The residue was reduced to dryness, brought to pH 4 with 1M HCl and extracted with EtOAc. The ethyl acetate solution was washed with H_2O (2x), brine (1x) and dried. Chromatography on silica gel (EtOAc/hexane 1:3 with 0.5 ml of AcOH per 100 ml of eluent) afforded the 1(10)-ene acid **36** (74 mg, 72%), the spectral data of which were fully in accord with those previously reported[36] for this compound.

The dimesylate **48** was prepared according to the protocol of Hanson[97] and subjected to the same Li/NH_3 conditions (the same molar ratios of the reagents were used) to afford, after chromatography, the 1(10)-ene acid **36** (78% yield).

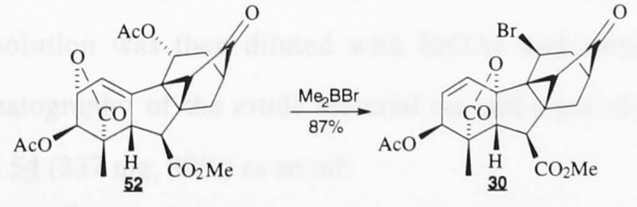
***ent*-1α,11α-Dibromo-2β-hydroxy-16-oxo-17,20-dinorgibberella-9-ene-7,19-dioic Acid 7-(Methyl ester) 19,2-Lactone (37)**



A solution of ketone **22** (14 mg, 0.042 mmol), which was prepared according to the protocol of Kraft-Klaunzer[37], N-bromosuccinimide (23 mg, 0.059 mmol, freshly crystallised) and dibenzoylperoxide (0.5 mg, 2 μmol) was heated at reflux for 30 minutes, whereupon TLC indicated that the starting material had been converted into a number of products. The reaction mixture was cooled, diluted with Et_2O and washed with a saturated aqueous solution of $Na_2S_2O_3$, $KHCO_3$ and brine. After drying and concentration under reduced pressure, chromatography on silica gel (EtOAc/hexane 2:3) afforded dibromoderivative **37** (1.3 mg, 6%) as an oil:

^{1}H NMR (300 MHz, $CDCl_3$) 5.11 (1H, d, $J_{11\alpha,12\alpha}$ = 6.2 Hz, H11α), 4.91 (1H, d, $J_{1\alpha,2\beta}$ = 3.8 Hz, H1), 4.8 (1H, dd, $J_{2\beta,1\alpha}$ = 3.8 Hz, $J_{2\beta,3\alpha}$ = 5.9 Hz, H2β), 3.78 (3H, s, -CO_2Me), 3.66 (1H, d, $J_{5,6}$ = 9.3 Hz, H5), 3.07 (1H, d, $J_{6,5}$ = 9.3 Hz, H6), 2.83 (1H, d, $J_{14\beta,14\alpha}$ = 13.0 Hz, H14β), 2.76 (1H, dd, $J_{15\beta,15\alpha}$ = 18.5 Hz, $J_{15\beta,14\alpha}$ = 3.7 Hz, H15β), 2.02 (1H, d, $J_{15\alpha,15\beta}$ = 18.5 Hz, H15α), 1.61 (1H, dd, $J_{14\alpha,14\beta}$ = 13.0 Hz, $J_{14\alpha,15\beta}$ = 3.7 Hz, H14α), 1.18 (3H, s, H18).

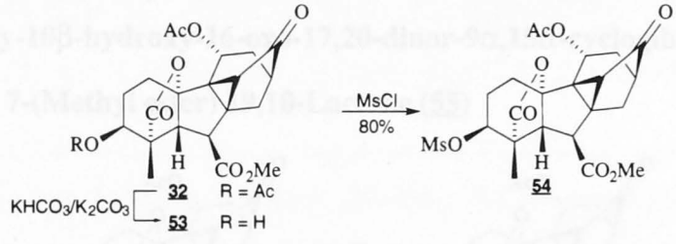
***ent*-3α-Acetoxy-11α-bromo-10β-hydroxy-16-oxo-17,20-dinor-9α,15α-cyclogibberella-1-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (30)**



1(10)-ene **52**[61] (350 mg, 0.788 mmol) was dissolved in dry CH_2Cl_2 (5 ml) under an argon atmosphere. The solution was cooled to -23°C in a CCl_4/dry ice bath and Me_2BBr (approximately 0.40 ml, 4.10 mmol) was added *via* a glass pipette. When

TLC indicated complete conversion into a more polar product, a 1M aqueous solution of $NaHCO_3$ was added and the resultant mixture was allowed to warm to room temperature. The mixture was diluted with EtOAc, the organic phase separated, washed with brine (2x), dried and the solvent removed. Chromatography on silica gel (EtOAc/hexane 1:1) afforded compound **30** (319 mg, 87%) as a crystalline solid, identical with the previously prepared sample[29].

***ent*-11β-Acetoxy-10β-hydroxy-3α-mesyloxy-16-oxo-17,20-dinor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (54)**



Diacetate **32**[27] (390 mg, 0.873 mmol) was dissolved in methanol (40 ml) and an aqueous solution of $KHCO_3/K_2CO_3$ was added (1 ml, 0.5M, 50 mg $KHCO_3$/70 mg K_2CO_3 in 1 ml of solution). The mixture was stirred at room temperature for two hours, at which stage TLC revealed that the reaction was complete. The solution was then poured into brine and the resultant mixture extracted with EtOAc. The organic layer was dried, solvent evaporated *in vacuo* and the crude alcohol **53** redissolved in CH_2Cl_2 (62 ml) and Et_3N (3.7 ml, 26.5 mmol). After cooling in an ice/water bath, methanesulfonyl chloride (0.65 ml, 8.4 mmol) was added dropwise, the mixture was allowed to gradually warm up to room temperature and stirring was continued for 14 hours. The solution was then diluted with EtOAc and subjected to standard work-up. Chromatography of the crude material on silica gel (EtOAc/hexane 1:1) afforded mesylate **54** (337 mg, 80%) as an oil:

$[\alpha]_D^{20}$ +17.8 (c 31 x 10^{-3}, CH_2Cl_2)

IR ($CDCl_3$) ν_{max} 2960, 1784, 1740, 1450, 1350, 1245, 1180, 1040 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 5.91 (1H, dd, $J_{11\beta,12\beta}$ = 9.5 Hz, $J_{11\beta,12\alpha}$ = 2.4 Hz, H11β), 4.70 (1H, m, H3α), 3.76 (3H, s, -$CO_2\underline{CH_3}$), 3.08 (3H, s, -$SO_3\underline{CH_3}$), 3.04 (1H, d,

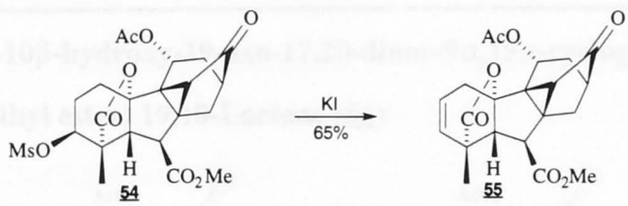
$J_{6,5}$ = 9.3 Hz, H6), 2.51 (1H, d, $J_{5,6}$ = 9.3 Hz, H5), 2.49-2.16 (6H, m), 2.12 (3H, s, $OCO\underline{CH_3}$), 1.98 (1H, d, J = 1.5 Hz, H15), 1.96-1.68 (3H, m), 1.20 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 209.5 (C16), 174.3 (CO), 171.0 (2xCO), 92.2 (C10), 78.4 (C3), 62.7 (C11), 52.8 ($-CO_2\mathbf{C}H_3$), 51.5 (C), 49.4 (CH), 46.9 (C), 46.2 (CH), 44.3 (C), 41.0 (CH), 38.9 ($-OSO_2\mathbf{C}H_3$), 35.6 (CH_2), 32.2 (CH), 28.6 (CH_2), 26.5 (CH_2), 24.1 (CH_2), 21.7 ($-OCO\mathbf{C}H_3$), 14.2 (C18);

LRMS 440 ($M^{\cdot+}$ - CH_2CO, 14), 422 ($M^{\cdot+}$ - AcOH, 17), 394 (3), 344 (8), 326 (5), 312 (4), 298 (19), 282 (38), 254 (36), 239(16), 223 (43), 195 (69), 181 (34), 55 (100);

HRMS found 422.1040 ($M^{\cdot+}$ - AcOH), $C_{20}H_{22}O_8S$ requires 422.1035.

***ent*-11β-Acetoxy-10β-hydroxy-16-oxo-17,20-dinor-9α,15α-cyclogibberella-2-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>55</u>)**



To a solution of methanesulfonyloxy derivative **<u>54</u>** (320 mg, 0.663 mmol) in dry DMF (32 ml) was added dry 18-crown-6 ether (320 mg, 1.21 mmol), dry KI (5.6 g, 33.7 mmol) and molecular sieves 4Å (8 g) and the reaction mixture was heated at 105°C for 48 hours. The mixture was diluted with EtOAc and subjected to standard work-up followed by washing with aqueous $CuSO_4$ (1x). The organic phase was dried, concentrated *in vacuo* and chromatographed on silica gel (EtOAc/hexane 2:3) to give the olefin **<u>55</u>** (0.167 g, 65 %), which crystallised from Et_2O/hexane as colourless needles:

mp 162-163°C;

$[\alpha]_D^{20}$ -114.3° (c 40.6 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2925, 1780, 1740, 1445, 1375, 1245, 1160, 1130, 1030, 1070 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 5.95 (1H, dd, $J_{11\beta,12\beta}$ = 9.7 Hz, $J_{11\beta,\ 12\alpha}$ = 2.5 Hz, H11β), 5.80 (1H, dt, $J_{2,3}$ = 9.3 Hz, $J_{2,1\alpha}$ = $J_{2,1\beta}$ = 3.4 Hz, H2), 5.62 (1H, dt, $J_{3,2}$ = 9.3 Hz, $J_{3,1\alpha}$ = $J_{3,1\beta}$ = 1.8 Hz, H3), 3.75 (3H, s, $-CO_2\underline{CH_3}$), 3.02 (1H, d,

$J_{6,5}$ = 9.3 Hz, H6), 2.76 (1H, ddd, $J_{1\alpha,1\beta}$ = 18.8 Hz, $J_{1\alpha,2}$ = 3.4 Hz, $J_{1\alpha,3}$ = 1.8 Hz, H1α), 2.48-2.26 (5H, m), 2.22 (1H, d, $J_{5,6}$ = 9.3 Hz, H5), 2.13 (3H, s, OCO<u>C</u>H$_3$), 1.88 (1H, J = 1.7 Hz, H15), 1.86 (1H, m overlapped), 1.24 (3H, s, H18);

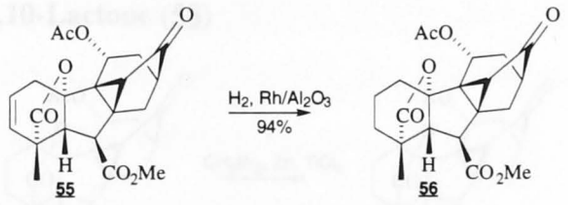
^{13}C NMR (75.5 MHz, $CDCl_3$) 210.0 (C16), 176.1 (CO), 171.2 (CO), 171.1 (CO), 132.3 (C2 or C3), 128.1 (C2 or C3), 90.7 (C10), 62.7 (C11), 52.9 (CH), 52.6 (-CO_2**C**H$_3$), 47.6 (C), 46.3 (CH), 45.9 (C), 45.4 (C), 41.0 (CH), 35.5 (CH_2), 32.2 (CH_2), 32.1 (CH), 28.8 (CH_2), 21.8 (-OCO**C**H$_3$), 14.6 (C18);

LRMS 386 ($M^{\cdot+}$, 1), 344 ($M^{\cdot+}$ - CH_2CO, 10), 327 (20), 312 (7), 295 (3), 282 (21), 267 (4), 250 (5), 240 (27), 223 (45), 208 (6), 195 (58), 180 (100), 165 (38);

HRMS found 344.1259 ($M^{\cdot+}$ - CH_2CO), $C_{19}H_{20}O_6$ requires 344.1260.

Anal. Found: C, 65.67; H, 6.02. Calcd for $C_{21}H_{22}O_7$: C, 65.28; H, 5.74.

***ent*-11β-Acetoxy-10β-hydroxy-19-oxo-17,20-dinor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>56</u>)**



Olefin **<u>55</u>** (110 mg, 0.285 mmol) was dissolved in EtOAc (11 ml). 5% Rh on alumina (20 mg) was added to the solution and the reaction mixture was stirred in the hydrogen atmosphere for 14 hours. Another portion of the catalyst was then added (40 mg) and stirring was continued for a further 5 hours. The mixture was diluted with EtOAc, the catalyst filtered off (using filter paper) and the solution concentrated *in vacuo*. Chromatography on silica gel (EtOAc/hexane 2:3) afforded the saturated derivative **<u>56</u>** (103 mg, 94%), which crystallised from ether/hexane:

mp 122-123°C;

$[\alpha]_D^{20}$ -13.4° (c 36.2 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2950, 1780, 1740, 1450, 1370, 1245, 1200, 1180, 1135, 1045 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 5.89 (1H, dd, $J_{11\beta,12\beta}$ = 9.6 Hz, $J_{11\beta,12\alpha}$ = 2.4 Hz, H11β), 3.75 (3H, s, -CO_2<u>C</u>H$_3$), 3.04 (1H, d, $J_{6,5}$ = 9.1 Hz, H6), 2.42 (1H, ddd,

J_1 = 11.8 Hz, J_2 = 5.6 Hz, J_3 = 1.8 Hz), 2.36-2.25 (3H, m), 2.23 (1H, d overlapped, $J_{14\alpha,\ 14\beta}$ = 12.0 Hz, H14α), 2.14 (3H, s, OCO<u>CH_3</u>), 1.97 (1H, d, $J_{5,6}$ = 9.1 Hz, H5), 1.87 (1H, d, J = 1.5 Hz, H15), 1.87-1.35 (6H, m), 1.09 (3H, s, H18);

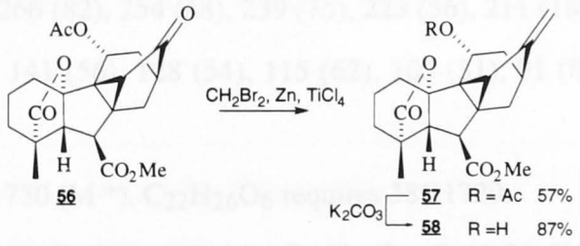
^{13}C NMR (75.5 MHz, $CDCl_3$) 210.2 (C16), 177.5 (CO), 171.5 (CO), 170.9 (CO), 91.8 (C10), 62.7 (C11), 56.0 (CH), 52.6 (-CO_2CH_3), 47.5 (C), 46.7 (CH), 46.6 (C), 44.4 (C), 41.0 (CH), 35.7 (CH_2), 34.8 (CH_2), 32.3 (CH), 28.6 (CH_2), 27.1 (CH_2), 21.7 (-OCOCH_3), 18.9 (CH_2), 16.5 (C18);

LRMS 388 ($M^{\cdot+}$, 12), 346 (38), 328 (38), 314 (18), 300 (62), 286 (21), 268 (23), 257 (17), 241 (47), 225 (50), 213 (12), 197 (48), 183 (19) 169 (13), 155 (19), 141 (32), 128 (30), 115 (32), 105 (21), 91 (55), 77 (44), 65 (29), 59 (43), 55 (100);

HRMS found 388.1522 ($M^{\cdot+}$), $C_{21}H_{24}O_7$ requires 388.1522.

Anal. Found: C, 65.06; H, 6.25. Calcd for $C_{21}H_{22}O_7$: C, 64.94; H, 6.23.

***ent*-10β,11β-Dihydroxy-20-nor-9α,15α-cyclogibberell-16-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>58</u>)**



To a stirred solution of activated zinc dust (1.0 g, 15.3 mmol) in dry THF (10 ml) and CH_2Br_2 (0.35 ml, 4.9 mmol) at -40°C, was added $TiCl_4$ (0.4 ml, 3.65 mmol), dropwise over 10 minutes[29]. The mixture was then stirred under N_2 at 4°C for 17 hours. The resulting suspension was added dropwise in 0.5 ml portions to the solution of ketone **<u>56</u>** (93 mg, 0.239 mmol) in dry CH_2Cl_2 (5 ml) until TLC indicated that the starting material had disappeared. The reaction was quenched with an aqueous $NaHCO_3$ solution, the resultant mixture stirred for 5 minutes, transferred into a separating funnel and extracted with Et_2O. The organic phase was washed with brine (2x), dried and concentrated *in vacuo*. Chromatography on silica gel

(EtOAc/hexane 3:7) afforded acetate **57** (53 mg, 57%), which crystallised from Et_2O/hexane:

mp 144-145°C;

$[\alpha]_D^{20}$ -10.2° (c 31.6 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2925, 1770, 1730, 1440, 1370, 1250, 1185, 1140, 1050, 1030 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 5.74 (1H, dd, $J_{11\beta,12\beta}$ = 10.1 Hz, $J_{11\beta,\ 12\alpha}$ = 3.4 Hz, H11β), 4.78 (2H, s, H17), 3.72 (3H, s, $-CO_2\underline{CH_3}$), 3.00 (1H, d, $J_{6,5}$ = 9.2 Hz, H6), 2.44 (1H, m), 2.29 (1H, m), 2.11 (3H, s, $OCO\underline{CH_3}$), 2.11-1.96 (4H, m), 1.89 (1H, d, $J_{14\alpha,\ 14\beta}$ = 11.5 Hz, H14α), 1.77 (1H, m), 1.66 (1H, m), 1.61-1.34 (4H, m), 1.07 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 178.2 (CO), 172.8 (CO), 171.3 (CO), 150.2 (C16), 103.3 (C17), 92.8 (C10), 64.9 (C11), 56.3 (CH), 52.2 ($-CO_2\mathbf{CH_3}$), 46.6 (C), 46.0 (CH), 42.1 (C), 41.6 (C), 37.5 (CH), 37.3 (CH_2), 34.9 (CH_2), 32.3 (CH_2), 30.1 (CH), 27.2 (CH_2), 21.9 ($-OCO\mathbf{CH_3}$), 19.1 (CH_2), 16.5 (C18);

LRMS 386 ($M^{\cdot+}$, 22), 344 ($M^{\cdot+}$ - CH_2CO, 1), 326 ($M^{\cdot+}$ - AcOH, 61), 312 (11), 298 (26), 282 (16), 266 (82), 254 (68), 239 (75), 223 (56), 211 (18), 195 (70), 181 (39), 167 (36), 155 (49), 141 (56), 128 (54), 115 (62), 105 (31), 91 (81), 77 (73), 59 (70), 55 (100);

HRMS found 386.1730 ($M^{\cdot+}$), $C_{22}H_{26}O_6$ requires 386.1729.

Anal. Found: C, 68.60; H, 6.94. Calcd for $C_{22}H_{26}O_6$: C, 68.38; H, 6.78.

The acetate (53 mg, 0.1317 mmol) was dissolved in MeOH (5 ml) and treated with an aqueous solution of $KHCO_3/K_2CO_3$ (0.5 ml, 0.5 M, 50 mg $KHCO_3$/70 mg K_2CO_3 in 1 ml of solution). More K_2CO_3 (25 mg, 0.181 mmol) was added at about 50% conversion and stirring was continued for another 48 hours. The reaction mixture was then diluted with EtOAc, washed with brine, dried and concentrated *in vacuo*. Chromatography on silica gel (EtOAc/hexane 2:3) afforded alcohol **58** (41 mg, 87%) as an oil:

$[\alpha]_D^{20}$ -53.4° (c 43.3 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 3590, 2960, 1780, 1730, 1660, 1460, 1440, 1380, 1340, 1280, 1250, 1200, 1175, 1120, 1070, 1030 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 4.75 (1H, s, H17), 4.73 (1H, s, H'17), 4.57 (1H, dt, $J_{11\beta,12\beta}$ = 8.7 Hz, $J_{11\beta,\ 12\alpha}$ = 1.93 Hz, $J_{11\beta,\ OH}$ = 1.93 Hz, H11β), 3.72 (3H, s, $-CO_2\underline{CH_3}$), 3.26 (1H, d, $J_{OH,11\beta}$ = 1.93 Hz, -OH), 3.01 (1H, d, $J_{6,5}$ = 9.1 Hz, H6), 2.44 (1H, m), 2.24 (1H, m), 2.14 (1H, d, $J_{5,6}$ = 9.1 Hz, H5), 2.02-1.44 (10H, m), 1.11 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 177.3 (CO), 172.7 (CO), 151.3 (C16), 102.3 (C17), 95.1 (C10), 64.5 (C11), 56.2 (CH), 52.3 ($-CO_2\mathbf{C}H_3$), 46.6 (C), 46.2 (CH), 42.9 (C), 42.0 (C), 39.6 (CH_2), 38.0 (CH), 35.2 (CH_2), 31.7 (CH_2), 30.6 (CH), 27.5 (CH_2), 19.1 (CH_2), 16.6 (C18);

LRMS 344 ($M^{\cdot+}$, 3), 326 ($M^{\cdot+}$ - H_2O, 3), 312 (48), 298 (9), 284 (24), 266 (10), 254 (61), 240 (81), 223 (23), 211 (21), 195 (88), 181 (36), 165 (33), 155 (52), 141 (69), 128 (61), 115 (68), 105 (32), 91 (80), 77 (85), 65 (54), 59 (100);

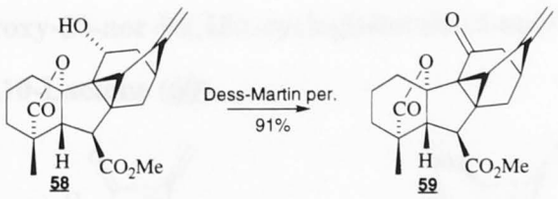
HRMS found 344.1622 ($M^{\cdot+}$), $C_{20}H_{24}O_5$ requires 344.1624.

Anal. Found: C, 69.40; H, 7.07. Calcd for $C_{20}H_{24}O_5$: C, 69.75; H, 7.02.

GC-MS (11-OTMS) 416 ($M^{\cdot+}$, 5), 401 (7), 384 (11), 356 (12), 341 (8), 326 (20), 313 (10), 281 (45), 266 (100), 254 (8), 241 (23), 223 (43);

KRI 2403.

***ent*-10β-Hydroxy-11-oxo-20-nor-9α,15α-cyclogibberell-16-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>59</u>)**



To the solution of alcohol **<u>58</u>** (22 mg, 0.064 mmol) in CH_2Cl_2 (2 ml) was added the Dess-Martin periodinane* (55 mg, 0.130 mmol) and the suspension was stirred for 14 hours at room temperature. The reaction mixture was then diluted with EtOAc, washed with aqueous $KHCO_3$ (1x), saturated aqueous $Na_2S_2O_3$ (1x), dried and

*The Dess-Martin periodinane referred to throughout the Experimental was prepared following the procedure of Ireland and Liu[64] and used freshly.

concentrated *in vacuo*. Chromatography on silica gel (EtOAc/hexane 2:3) gave ketone **59** (20 mg, 91%), which crystallised from Et_2O/hexane:

mp 199-200°C;

$[\alpha]_D^{20}$ -113.0° (c 28.0 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2960, 1770, 1735, 1700, 1460, 1380, 1300, 1280, 1260, 1200, 1175, 1125, 1055, 995 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 4.96 (1H, s, H17), 4.94 (1H, s, H'17), 3.75 (3H, s, $-CO_2\underline{CH_3}$), 3.02 (1H, d, $J_{6,5}$ = 9.1 Hz, H6), 2.69 (1H, m), 2.66 (1H, m overlapped), 2.61 (1H, s, H15), 2.25 (1H, dd, $J_{14\beta,14\alpha}$ = 12.0 Hz, $J_{14\beta,13}$ = 5.6 Hz, H14β), 2.18 (1H, s, H12), 2.17 (1H, s, H'12), 2.12 (1H, d, $J_{5,6}$ = 9.1 Hz, H5), 1.84 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.0 Hz, H14α), 1.91-1.39 (6H, m), 1.10 (3H, s, H18);

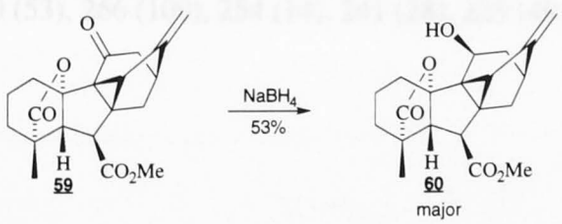
^{13}C NMR (75.5 MHz, $CDCl_3$) 202.3 (C11), 178.2 (CO), 172.0 (CO), 147.5 (C16), 106.2 (C17), 90.4 (C10), 55.8 (CH), 52.7 (CH_2), 52.6 ($-CO_2\mathbf{C}H_3$), 47.2 (C), 45.7 (CH), 45.6 (C), 44.5 (C), 36.5 (CH), 34.8 (CH_2 + CH), 33.6 (CH_2), 28.3 (CH_2), 19.1 (CH_2), 16.7 (C18);

LRMS 342 ($M^{\cdot+}$, 67), 298 (28), 283 (25), 270 (17), 266 (33), 261 (4), 255 (37), 239 (100), 223 (25), 211 (65), 195 (55), 181 (40);

HRMS found 342.1469 ($M^{\cdot+}$), $C_{20}H_{22}O_5$ requires 342.1467.

Anal. Found: C, 69.91; H, 6.77. Calcd for $C_{20}H_{22}O_5$: C, 70.16; H, 6.48.

***ent*-10β,11α-Dihydroxy-20-nor-9α,15α-cyclogibberell-16-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (60)**



The solution of ketone **59** (19 mg, 0.055 mmol) in dimethoxyethane (1 ml) was treated with $NaBH_4$ (4 mg, 0.106 mmol) at room temperature. TLC indicated complete conversion into a mixture of two products in 5 minutes. The reaction mixture was

diluted with EtOAc, subjected to standard work-up, the organic phase dried and concentrated *in vacuo*. Chromatography on silica gel (EtOAc/hexane 2:3) afforded 11α-alcohol **58** (5 mg, 26%), identical in all respects with the previously prepared material, followed by the more polar 11β-alcohol **60** (10 mg, 53%), which was obtained as an oil:

$[\alpha]_D^{20}$ -72.1° (c 24.0 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 3600, 2925, 1765, 1730, 1665, 1460, 1435, 1280, 1200, 1175, 1135, 1045, 990 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 4.90 (1H, s, H17), 4.87 (1H, s, H'17), 4.52 (1H, bt, J = 8.6 Hz, H11α), 3.73 (3H, s, $-CO_2\underline{CH_3}$), 2.92 (1H, d, $J_{6,5}$ = 9.0 Hz, H6), 2.49 (2H, m), 2.14 (1H, s overlapped, H15), 2.12 (1H, d, $J_{5,6}$ = 9.0 Hz, H5), 1.98 (2H, m), 1.86-1.41 (8H, m), 1.10 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 178.8 (CO), 172.8 (CO), 150.6 (C16), 104.0 (C17), 93.3 (C10), 65.2 (C11), 55.6 (C5), 52.3 ($-CO_2\mathbf{C}H_3$), 48.0 (C), 46.4 (C), 46.3 (C6), 43.5 (C), 42.1 (C12), 38.0 (C13), 35.1 (C3 or C14), 33.7 (C3 or C14), 29.0 (C1), 28.9 (C15), 19.3 (C2), 16.8 (C18);

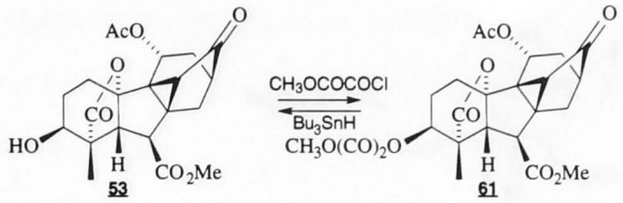
LRMS 344 ($M^{\cdot+}$, 3), 326 ($M^{\cdot+}$ - H_2O, 3), 312 (37), 300 (8), 284 (20), 266 (8), 254 (40), 240 (56), 223 (17), 211 (16), 197 (64), 181 (29), 167 (26), 155 (40),149 (26), 141 (49), 128 (46), 115 (54), 105 (29), 97 (17), 91 (69), 77 (76), 65 (47), 55 (100);

HRMS found 344.1622 ($M^{\cdot+}$), $C_{20}H_{24}O_5$ requires 344.1624.

GC-MS (11-OTMS) 416 ($M^{\cdot+}$, 10), 400 (3), 384 (11), 372 (7), 356 (11), 340 (5), 326 (24), 311 (9), 280 (53), 266 (100), 254 (14), 241 (28), 223 (49);

KRI 2413.

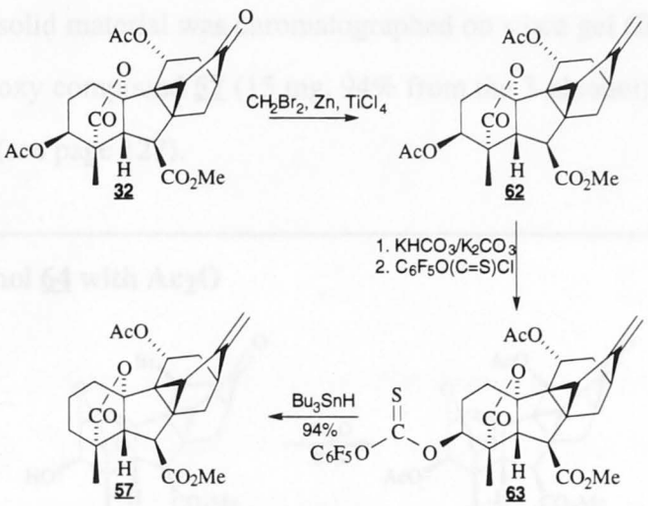
Attempted removal of the 3-hydroxy group by the deoxygenation of the 3-methyloxalyloxy derivative 61



Alcohol **53** (crude, 30 mg, 0.074 mmol) obtained by the hydrolysis of diacetate **32** (see page 124) was dissolved in dry CH_2Cl_2 (1 ml). Et_3N (20 µl, 0.143 mmol) and $CH_3OCOCOCl$ (20 µl, 0.217 mmol) were added to the solution, together with a catalytic amount of DMAP. After 16 hours at room temperature, the reaction mixture was diluted with EtOAc, subjected to standard work-up, the organic phase dried and the solvent removed. The residue contained 3-methyloxalyl compound **61** (33 mg, 92% crude).

The crude methyloxalyl derivative **61** (20 mg, 0.041 mmol) was, without chracterisation, dissolved in Bu_3SnH (0.2 ml), the solution was heated at 110°C and the initiator (AIBN) was added at this temperature. The reaction, which was complete within a few minutes (as judged by TLC analysis), afforded a sole product. Hexane was then added to the solution, which resulted in the formation of a white precipitate. The liquid was removed and the vial containing the solid material was rinsed with hexane (3 x 0.5 ml) in order to remove the tin compounds. Chromatography of the residue on silica gel (EtOAc/hexane 3:2) afforded the parent alcohol **53** (16 mg), identical with the previously prepared material.

Removal of the 3-hydroxy group *via* the pentafluorophenoxythionoformate derivative 63



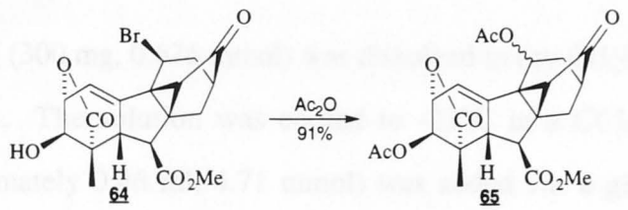
Olefin **62** was prepared[27] from ketone **32** *via* the Lombardo-Oshima methylenation. The hydrolysis of the 3-acetate group was carried out following the same procedure as described for the hydrolysis of the corresponding 3-acetoxy group in ketone **32** (the same molar ratios and concentration of the substrate were used, see page 124).

The crude alcohol (17 mg, 0.042 mmol) was dissolved in CH_2Cl_2 (1 ml) and the following reagents were added: dry Et_3N (20 µl, 0.143 mmol), DMAP (20 mg, 0.164 mmol) and pentafluorophenoxychlorothionoformate (20 µl, 0.125 mmol). The solution was allowed to stand at room temperature with the exclusion of light. TLC analysis indicated that the esterification was complete over two hours. The reaction mixture was diluted with EtOAc, subjected to standard work-up, the organic solution dried and solvent removed (on a rotary evaporator then under high vacuum). It is important to note that it was desirable to work with the exclusion of direct light and reduce the time needed to carry out these operations to minimum, due to the observed sensitivity of these esters towards light[67]. However, it was found by others[67] that if required, these compounds could be purified by column chromatography provided that the aforementioned precautions are taken.

The residue was dissolved in dry benzene (1.0 ml) and Bu_3SnH (0.1 ml, 0.372 mmol) was added to the solution. The reaction mixture was heated at reflux for

30 minutes and AIBN (catalytic amount) was added twice at 15-minute intervals. The solvent was then removed and the residue was kept under high vacuum at 80°C for 30 minutes. The solid material was chromatographed on silica gel (EtOAc/hexane 2:3) to afford the 3-deoxy compound **57** (15 mg, 94% from the 3-alcohol), identical with an authentic sample (see page 127).

Reaction of alcohol 64 with Ac_2O



Alcohol **64** (obtained by accidental hydrolysis of the 3-acetoxy group in **29**, approximately 400 mg, 0.862 mmol) was dissolved in CH_2Cl_2 (5 ml). Et_3N (0.7 ml, 5.02 mmol) and Ac_2O (0.5 ml, 5.30 mmol) were added and the reaction mixture was allowed to stand overnight. The solution was diluted with EtOAc, subjected to standard work-up, dried and the solvent removed. Chromatography on silica gel (EtOAc/hexane 2:3) afforded the mixture of epimeric acetates **65** (11β:11α = 2:1, 350 mg, 91%) as an oil:

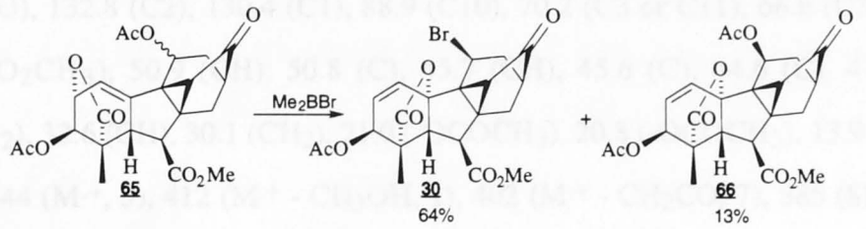
^{1}H NMR (300 MHz, $CDCl_3$) 5.86 (dd, $J_{1,2\beta}$ = 5.9 Hz, $J_{1,5}$ = 2.7 Hz, H1), 5.50 (1H, d, $J_{11\alpha,12\alpha}$ = 8.0 Hz, H11α), 4.97 (m, H2β, H3α), 3.79 (3H, s, -$CO_2\underline{CH_3}$), 3.02 (d, $J_{6,5}$ = 9.8 Hz, H6), 2.92 (dd overlapped, $J_{5,6}$ = 9.8 Hz, $J_{5,1}$ = 2.7 Hz, H5), 2.13 (s, -OCO$\underline{CH_3}$), 2.06 (s, -OCO$\underline{CH_3}$), 1.18 (s, H18) (**major epimer**);

^{1}H NMR (300 MHz, $CDCl_3$) 5.77 (m, H1, H11β), 4.97 (m, H2β, H3α), 3.79 (3H, s, -$CO_2\underline{CH_3}$), 3.07 (d, $J_{6,5}$ = 9.7 Hz, H6), 3.93 (dd overlapped, $J_{5,6}$ = 9.7 Hz, $J_{5,1}$ = 2.6 Hz, H5), 2.10 (s, -OCO$\underline{CH_3}$), 2.04 (s, -OCO$\underline{CH_3}$), 1.18 (s, H18) (**minor epimer**, these data being consistent with those for compound **52**[27,61]);

LRMS 444 ($M^{\cdot+}$, 13), 412 ($M^{\cdot+}$ - CH_3OH, 5), 402 ($M^{\cdot+}$ - CH_2CO, 17), 384 (5), 356 (4), 339 (7), 325 (35), 298 (74), 281 (15), 270 (13), 254 (36), 239 (51), 221 (25), 211 (32), 195 (100) (**mixture**);

HRMS found 444.1419 ($M^{\cdot+}$), $C_{23}H_{24}O_9$ requires 444.1420.

Reaction of mixture 65 with Me_2BBr



Mixture **65** (300 mg, 0.676 mmol) was dissolved in dry CH_2Cl_2 (5 ml) under an argon atmosphere. The solution was cooled to -23°C in a CCl_4/dry ice bath and Me_2BBr (approximately 0.46 ml, 4.71 mmol) was added *via* a glass pipette. After 4 hours at -23°C, 1M aqueous solution of $NaHCO_3$ was added and the resultant mixture was allowed to warm to room temperature. The mixture was diluted with EtOAc, the organic phase separated, washed with brine (2x), dried and the solvent removed. Chromatography on silica gel (EtOAc/hexane 1:1) afforded in order of elution:

***ent*-3α-Acetoxy-11α-bromo-10β-hydroxy-16-oxo-17,20-dinor-9α,15α-cyclogibberella-1-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**30**, 200 mg, 64%);

***ent*-3α,11α-Diacetoxy-10β-hydroxy-16-oxo-17,20-dinor-9α,15α-cyclogibberella-1-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**66**, 40 mg, 13%) as an oil:

^{1}H NMR (300 MHz, $CDCl_3$) 6.61 (1H, d, $J_{1,2}$ = 9.4 Hz, H1), 5.85 (1H, dd, $J_{2,1}$ = 9.4 Hz, $J_{2,3\alpha}$ = 3.8 Hz, H2), 5.61 (1H, dd, $J_{11\alpha,12\alpha}$ = 8.4 Hz, $J_{11\alpha,12\beta}$ = 2.0 Hz, H11α), 5.37 (1H, d, $J_{3\alpha,2}$ = 3.8 Hz, H3α), 3.78 (3H, s, -$CO_2\underline{CH_3}$), 3.04 (1H, d, $J_{6,5}$ = 9.4 Hz, H6), 2.81 (1H, d, $J_{5,6}$ = 9.4 Hz, H5), 2.45 (1H, ddd overlapped, $J_{12\alpha,12\beta}$ = 15.3 Hz, $J_{12\alpha,11\alpha}$ = 8.4 Hz, $J_{12\alpha,13}$ = 2.9 Hz, H12α), 2.41 (1H, ddd overlapped, $J_{14\beta,14\alpha}$ = 11.7 Hz, $J_{14\beta,13}$ = 5.7 Hz, J = 1.6 Hz, H14β), 2.34 (1H, m, H13), 2.27 (1H, s, H15), 2.12 (3H, s, -OCO$\underline{CH_3}$), 2.07 (3H, s, -OCO$\underline{CH_3}$), 1.84 (1H,

d overlapped, $J_{14\alpha,14\beta}$ = 11.7 Hz, H14α), 1.80 (1H, dm overlapped, $J_{12\beta,12\alpha}$ = 15.3 Hz, H12β), 1.20 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 208.8 (C16), 175.7 (CO), 170.8 (CO), 169.8 (CO), 169.7 (CO), 132.8 (C2), 130.4 (C1), 88.9 (C10), 70.2 (C3 or C11), 66.6 (C3 or C11), 52.7 (-CO_2CH_3), 50.9 (CH), 50.8 (C), 45.7 (CH), 45.6 (C), 44.6 (C), 41.4 (CH), 38.0 (CH_2), 32.6 (CH), 30.1 (CH_2), 21.0 (-OCOCH_3), 20.8 (-OCOCH_3), 13.9 (C18);

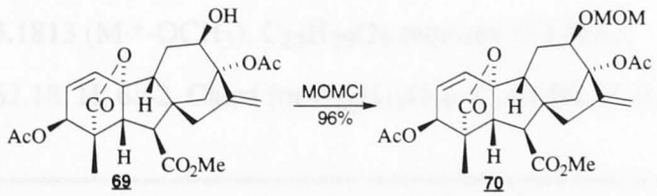
LRMS 444 ($M^{\cdot+}$, 3), 412 ($M^{\cdot+}$ - CH_3OH, 1), 402 ($M^{\cdot+}$ - CH_2CO, 7), 385 (8), 340 (8), 325 (10), 298 (100), 281 (14), 254 (24), 239 (33), 221 (21), 195 (44);

HRMS found 444.1419 ($M^{\cdot+}$), $C_{23}H_{24}O_9$ requires 444.1420.

5.3 CHAPTER 3 EXPERIMENTAL

5.3.1 Transannular oxidation pathway

***ent*-3α,13-Diacetoxy-10β-hydroxy-12α-methoxymethoxy-20-norgibberella-1,16-diene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (70)**



Alcohol **69**[44] (770 mg, 1.67 mmol) was dissolved in dry CH_2Cl_2 (20 ml). DIPEA (3 ml, 17.22 mmol) and DMAP (30 mg, 0.245 mmol) were added followed by MOMCl (1.2 ml, 15.80 mmol). The solution was left overnight, whereupon the reaction was judged to be complete by TLC. The mixture was diluted with EtOAc, subjected to standard work-up, dried and the solvent removed under reduced pressure. Chromatography on silica gel (EtOAc/hexane 3:7) gave derivative **70** (810 mg, 96%) as an oil, which crystallised from Et_2O/hexane:

mp 126-127°C;

$[\alpha]_D^{20}$ 147.0° (c 30.9 x 10^{-3}, EtOH);

IR ($CDCl_3$) ν_{max} 2960, 1780, 1730, 1440, 1370, 1250, 1040 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 6.38 (1H, d, $J_{1,2}$ = 9.3 Hz, H1), 5.88 (1H, dd, $J_{2,1}$ = 9.3 Hz, $J_{2,3\alpha}$ = 3.7 Hz, H2), 5.34 (1H, d, $J_{3\alpha,2}$ = 3.7 Hz, H3α), 5.25 (1H, bs, H17), 5.23 (1H, bs , H'17), 4.72 (1H, d, J = 6.8 Hz, -O$\underline{CH_2}$OCH$_3$), 4.69 (1H, d, J = 6.8 Hz, -O$\underline{CH_2}$OCH$_3$), 4.47 (1H, dd, $J_{12\alpha,11\alpha}$ = 7.2 Hz, $J_{12\alpha,11\beta}$ = 2.3 Hz, H12α), 3.76 (3H, s, -CO$_2\underline{CH_3}$), 3.40 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.35 (1H, d, $J_{5,6}$ = 10.6 Hz, H5), 2.76 (1H, d, $J_{6,5}$ = 10.6 Hz, H6), 2.43 (1H, d, $J_{15,15}$ = 11.1 Hz, H15), 2.13 (3H, s, -OCO$\underline{CH_3}$), 2.05 (3H, s, -OCO$\underline{C\ H_3}$), 1.77 (1H, ddd, $J_{11\beta,11\alpha}$ = 14.7 Hz, $J_{11\beta,9}$ = 6.5 Hz, $J_{11\beta,12\alpha}$ = 2.3Hz, H11β), 1.15 (3H, s, H18);

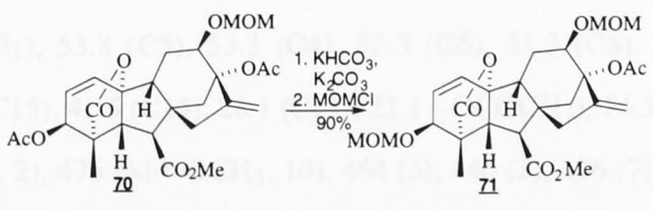
^{13}C NMR (75.5 MHz, $CDCl_3$) 176.9 (CO), 172.2 (CO), 170.0 (CO), 169.7 (CO), 144.2 (C16), 133.9 (C2), 129.3 (C1), 112.7 (C17), 95.5 (-O**C**H$_2$OCH$_3$), 90.0 (C10), 87.2 (C13), 75.9 (C12), 70.2 (C3), 55.5 (-OCH$_2$O**C**H$_3$), 53.8 (C5), 52.3 (C6), 52.1 (C4), 51.2 (C8), 50.0 (-CO$_2$**C**H$_3$), 48.7 (C9), 42.5 (C15), 41.3 (C14), 26.0 (C11), 22.1 (-OCO**C**H$_3$), 20.9 (-OCO**C**H$_3$), 14.3 (C18);

LRMS 473 (M·$^+$-OMe, 31), 462 (10), 444 (3), 434 (10), 410 (2), 401 (6), 370 (3), 358 (8), 339 (11), 325 918), 308 (13) 295 (33), 267 (83), 253 924), 235 (38), 223 925), 209 (100);

HRMS found 473.1813 (M·$^+$-OCH$_3$), $C_{25}H_{29}O_9$ requires 473.1812.

Anal. Found: C, 62.18; H, 6.42. Calcd for $C_{26}H_{32}O_{10}$: C, 61.90; H, 6.39.

***ent*-13-Acetoxy-10β-hydroxy-3α,12α-bis(methoxymethoxy)-20-norgibberella-1,16-diene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>71</u>)**



Diacetate **<u>70</u>** (800 mg, 1.59 mmol) was dissolved in methanol (38 ml) and an aqueous solution of $KHCO_3/K_2CO_3$ was added dropwise (0.95 ml, 0.5 M, 50 mg $KHCO_3$/70 mg K_2CO_3 in 1 ml of solution). The mixture was stirred at 0°C until TLC analysis revealed that the reaction was complete. The solution was then poured into

brine and the resultant mixture extracted with EtOAc. The organic layer was dried, solvent evaporated *in vacuo* and the crude alcohol redissolved in CH_2Cl_2 (19 ml) and DIPEA (3 ml). MOMCl (5 ml, 18.43 mmol) was added to the solution followed by DMAP (30 mg, 0.246 mmol). TLC analysis indicated that the starting material was consumed in 72 hours. The solution was then diluted with EtOAc, subjected to standard work-up, dried and the solvent evaporated. Chromatography of the residue (EtOAc/hexane 3:7) afforded the desired 3-methoxymethoxy compound **71** (721 mg, 90%) as an oil, which crystallised from Et_2O/hexane :

mp 108-109°C;

$[\alpha]_D^{20}$ 113.0° (c 28.6 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2960, 1770, 1730, 1570, 1440, 1370, 1240, 1150, 1100, 1030 cm^{-1};

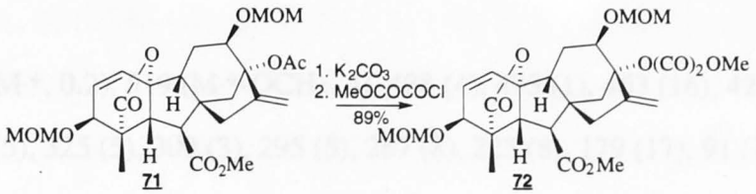
^{1}H NMR (300 MHz, $CDCl_3$) 6.29 (1H, d, $J_{1,2}$ = 9.4 Hz, H1), 5.94 (1H, dd, $J_{2,1}$ = 9.4 Hz, $J_{2,3\alpha}$ = 3.7 Hz, H2), 5.24 (1H, bs, H17), 5.22 (1H, bs , H'17), 4.74 (1H, d, J = 7.0 Hz, 3-O$\underline{CH_2}$OCH$_3$), 4.70 (2H, s overlapped, 12-O$\underline{CH_2}$OCH$_3$), 4.68 (1H, d overlapped, J = 7.0 Hz, 3-O$\underline{CH_2}$OCH$_3$), 4.46 (1H, dd, $J_{12\alpha,11\alpha}$ = 7.2 Hz, $J_{12\alpha,11\beta}$ = 2.4 Hz, H12α), 4.01 (1H, d, $J_{3\alpha,2}$ = 3.7 Hz, H3α), 3.74 (3H, s, -CO$_2\underline{CH_3}$), 3.40 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.38 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.33 (1H, d, $J_{5,6}$ = 10.6 Hz, H5), 2.75 (1H, d, $J_{6,5}$ = 10.6 Hz, H6), 2.41 (1H, d, $J_{15,15}$ = 11.2 Hz, H15), 2.04 (3H, s, -OCO$\underline{CH_3}$), 1.75 (1H, ddd, $J_{11\beta,11\alpha}$ = 14.7 Hz, $J_{11\beta,9}$ = 6.7 Hz, $J_{11\beta,12\alpha}$ = 2.4 Hz, H11β), 1.23 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 178.1 (CO), 172.3 (CO), 169.7 (CO), 144.4 (C16), 132.2 (C2), 131.5 (C1), 112.5 (C17), 97.0 (-O**C**H$_2$OCH$_3$), 95.7 (-O**C**H$_2$OCH$_3$), 90.4 (C10), 87.3 (C13), 76.1 (C12), 75.0 (C3), 55.9 (-OCH$_2$O**C**H$_3$), 55.5 (-OCH$_2$O**C**H$_3$), 53.8 (C5), 53.3 (C4), 52.3 (C6), 51.3 (C8), 50.3 (-CO$_2$**C**H$_3$), 48.8 (C9), 42.5 (C15), 41.5 (C14), 26.1 (C11), 22.1 (-OCO**C**H$_3$), 14.3 (C18);

LRMS 506 ($M^{\cdot+}$, 2), 475 ($M^{\cdot+}$-OCH_3, 10), 464 (3), 446 (2), 436 (7), 420 (6), 403 (7), 388 (5), 375 (11), 358 (11), 340 (12), 325 (18), 313 (16), 295 (24), 267 (66), 209 (73), 113 (100);

HRMS found 475.1968 ($M^{\cdot+}$-OCH_3), $C_{25}H_{31}O_9$ requires 475.1968.

***ent*-10β-Hydroxy-3α,12α-bis(methoxymethoxy)-13-methyloxalyloxy-20-norgibberella-1,16-diene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (72)**



Derivative **71** (720 mg, 1.42 mmol) was dissolved in MeOH (40 ml) and the solution was treated with 1M aqueous K_2CO_3 (1 ml). Another portion of the reagent (1 ml) was added after 5 hours at room temperature and the reaction mixture was stirred overnight. The solution was then poured into brine and the resultant mixture extracted with EtOAc. The organic layer was dried, solvent evaporated *in vacuo* and the crude alcohol redissolved in CH_2Cl_2 (15 ml) and Et_3N (1 ml). Methyloxalyl chloride (0.3 ml, 3.26 mmol) was then added dropwise followed by DMAP (20 mg, 0.164 mmol) and the reaction mixture was left overnight. The solution was diluted with EtOAc and subjected to standard work-up. The organic phase was dried, concentrated under reduced pressure and the solution chromatographed on silica gel (EtOAc/hexane 1:1) to afford the methyloxalyl derivative **72** (693 mg, 89%) as an oil:

$[\alpha]_D^{20}$ 114.0° (c 25.8 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2960, 1770, 1740, 1440, 1380, 1200, 1160, 1100, 1040 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 6.29 (1H, d, $J_{1,2}$ = 9.4 Hz, H1), 5.95 (1H, dd, $J_{2,1}$ = 9.4 Hz, $J_{2,3\alpha}$ = 3.7 Hz, H2), 5.29 (2H, bs, H17, H'17), 4.72 (4H, 2 AB systems overlapped, 3- and 12-O$\underline{CH_2}$OCH$_3$), 4.49 (1H, dd, $J_{12\alpha,11\alpha}$ = 7.3 Hz, $J_{12\alpha,11\beta}$ = 2.7 Hz, H12α), 4.00 (1H, d, $J_{3\alpha,2}$ = 3.7 Hz, H3α), 3.88 (3H, s, -O(CO)$_2$O$\underline{CH_3}$), 3.74 (3H, s, -CO$_2\underline{CH_3}$), 3.40 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.38 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.33 (1H, d, $J_{5,6}$ = 10.5 Hz, H5), 2.76 (1H, d, $J_{6,5}$ = 10.5 Hz, H6), 2.53 (1H, d, $J_{15,15}$ = 11.2 Hz, H15), 1.78 (1H, ddd, $J_{11\beta,11\alpha}$ = 14.7 Hz, $J_{11\beta,9}$ = 6.9 Hz, $J_{11\beta,12\alpha}$ = 2.7Hz, H11β), 1.23 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 178.0 (CO), 172.1 (CO), 158.0 (CO), 155.9 (CO), 142.5 (C16), 132.0 (C2), 131.7 (C1), 113.9 (C17), 96.9 (-O**C**H$_2$OCH$_3$), 95.7 (-OCH$_2$O**C**H$_3$), 90.2 (C10), 89.9 (C13), 75.7 (C12), 75.0 (C3),

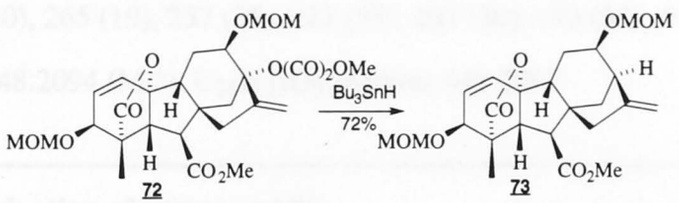
55.9 (-OCH_2OCH_3), 55.7 (-OCH_2OCH_3), 53.8 (C5), 53.6 (-$O(CO)_2CH_3$), 53.3 (C4), 52.3 (C6), 51.5 (C8), 50.0 (-CO_2CH_3), 48.7 (C9), 42.4 (C15), 40.8 (C14), 26.2 (C11), 14.6 (C18);

LRMS 550 ($M^{\cdot+}$, 0.2), 519 ($M^{\cdot+}$-OCH_3, 4), 488 (4), 473 (1), 463 (16), 429 (1), 385 (2), 357 (8), 339 (5), 325 (5), 309 (3), 295 (5), 267 (8), 223 (8), 179 (17), 91 (18), 59 (100);

HRMS found 519.1865 ($M^{\cdot+}$-OCH_3), $C_{26}H_{31}O_{11}$ requires 519.1866.

Anal. Found: C, 59.68; H, 7.02. Calcd for $C_{27}H_{34}O_{12}$: C, 58.90; H, 6.22.

***ent*-10β-Hydroxy-3α,12α-bis(methoxymethoxy)-20-norgibberella-1,16-diene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>73</u>)**



Methyloxalyl ester **<u>72</u>** (690 mg, 1.25 mmol) was dissolved in dry toluene (33 ml) under an argon atmosphere. Bu_3SnH (0.725 ml, 2.69 mmol) was added and the solution was heated at reflux. The reaction mixture was maintained at this temperature for two hours while catalytic amounts of AIBN were added at 30-minute intervals. More reagent (0.4 ml, 1.49 mmol) was added after this period and the reaction was run for another hour, at which stage TLC analysis indicated that the starting material completely disappeared and was replaced with a compound of lower R_f. The solvent was evaporated under reduced pressure and the excess of the reagent was removed under high vacuum at 80°C. Chromatography on silica gel (EtOAc/hexane 2:3) gave the 13-deoxy compound **<u>73</u>** (407 mg, 72%) as an oil:

$[\alpha]_D^{20}$ 108.6° (c 32.4 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2960, 1770, 1730, 1660, 1600, 1440, 1380, 1150, 1100, 1030 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 6.34 (1H, d, $J_{1,2}$ = 9.3 Hz, H1), 5.94 (1H, dd, $J_{2,1}$ = 9.3 Hz, $J_{2,3\alpha}$ = 3.7 Hz, H2), 5.08 (1H, bs, H17), 5.04 (1H, bs, H'17), 4.74 (1H, d, J = 7.0 Hz, 3-O<u>C</u>$H_2$$OCH_3$), 4.70 (1H, d, J = 6.7 Hz, 12-O<u>C</u>$H_2$$OCH_3$), 4.67 (1H, d, J = 7.0 Hz, 3-O<u>C</u>$H_2$$OCH_3$), 4.64 (1H, d, J = 6.7 Hz, 12-O<u>C</u>$H_2$$OCH_3$), 4.20 (1H,

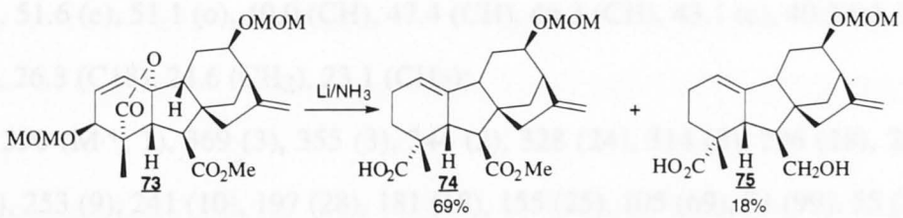
m overlapped, H12α), 4.20 (1H, d overlapped, $J_{3\alpha,2}$ = 3.7 Hz, H3α), 3.72 (3H, s, -$CO_2\underline{CH_3}$), 3.38 (3H, s, -$OCH_2O\underline{CH_3}$), 3.37 (3H, s, -$OCH_2O\underline{CH_3}$), 3.30 (1H, d, $J_{5,6}$ = 9.4 Hz, H5), 2.86 (1H, m, H13), 2.72 (1H, d, $J_{6,5}$ = 9.4 Hz, H6), 2.08 (1H, dd overlapped, J_1 = 14.4 Hz, J_2 = 7.1 Hz), 1.25 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 178.5 (CO), 173.1 (CO), 146.9 (C16), 132.1 (C2), 131.4 (C1), 111.1 (C17), 96.9 (-OCH_2OCH_3), 94.6 (-OCH_2OCH_3), 90.9 (C10), 75.4 (C12), 74.7 (C3), 55.8 (-OCH_2OCH_3), 55.4 (-OCH_2OCH_3), 54.3 (C5), 53.1 (C4), 52.8 (C8), 52.0 (C6), 49.8 (-CO_2CH_3), 49.1 (C9), 43.9 (C15), 43.8 (C13), 39.9 (C14), 24.5 (C11), 14.5 (C18);

LRMS 448 ($M^{\cdot+}$, 0.3), 417 ($M^{\cdot+}$-OCH_3, 5), 386 (31), 354 (7), 341 (6), 310 (10), 297 (18), 283 (20), 265 (19), 237 (25), 221 (35), 207 (30), 193 (58), 119 (62), 91 (100);

HRMS found 448.2094 ($M^{\cdot+}$), $C_{24}H_{32}O_8$ requires 448.2097.

Li/ammonia reduction of compound <u>73</u>



Compound <u>73</u> (405 mg, 0.90 mmol) was dissolved in dry THF (3.5 ml) together with t-BuOH (0.38 ml, 4.03 mmol). After cooling to -78°C in an acetone/dry ice bath, liquid ammonia (approximately 11 ml) was distilled into the flask from a $NaNH_2$ solution. Lithium metal (approximately 29 mg, 4.18 mmol) was then added in small pieces with efficient stirring and the consumption of the starting material was carefully monitored by TLC. Regrettably, too large an amount of Li metal was inadvertently added towards the end of the reduction, as indicated by the persistence of a faint blue colour, and as a consequence, some reduction of the 7-methyl ester function occurred as well. The reaction was immediately quenched with solid NH_4Cl and the ammonia allowed to evaporate. The residue was reduced to dryness, brought to pH 4 with 1M HCl and extracted with EtOAc. The ethyl acetate solution was washed with H_2O (2x),

brine (1x), dried and concentrated under reduced pressure. Chromatography on silica gel (EtOAc/hexane 2:3 with 1 ml of AcOH per 100 ml of eluent) afforded in order of elution:

***ent*-12α-Methoxymethoxy-20-norgibberella-1(10),16-diene-7,19-dioic Acid 7-(Methyl ester)** (**74**, 243 mg, 69%, oil):

IR ($CDCl_3$) ν_{max} 2960, 1720, 1690, 1650, 1460, 1450, 1440, 1360, 1150, 1100, 1050 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 5.41 (1H, m, H1), 4.99 (1H, bs, H17), 4.97 (1H, bs, H'17), 4.72 (1H, d, J = 6.9 Hz, -O$\underline{CH_2}$OCH$_3$), 4.63 (1H, d, J = 6.9 Hz, -O$\underline{CH_2}$OCH$_3$), 3.70 (3H, s, -CO$_2\underline{CH_3}$), 3.65 (1H, m, H12α), 3.38 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.04 (1H, d, $J_{6,5}$ = 5.3 Hz, H6), 2.86 (1H, m, H13), 2.65 (2H, m), 2.41 (2H, m), 1.32 (1H, ddd, $J_{3\alpha,3\beta}$ = 11.8 Hz, $J_{3\alpha,2\alpha}$ = 6.3 Hz, $J_{3\alpha,2\beta}$ = 1.7 Hz, H3β), 1.24 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 181.5 (CO), 176.7 (CO), 148.2 (C16), 140.4 (C10), 114.0 (C1), 108.7 (C17), 94.4 (-O**C**H$_2$OCH$_3$), 75.4 (C12), 55.3 (-OCH$_2$O**C**H$_3$), 51.6 (o), 51.6 (e), 51.1 (o), 49.0 (CH), 47.4 (CH), 46.3 (CH), 43.1 (e), 40.3 (e), 39.6 (e), 34.2 (e), 26.3 (C18), 24.6 (CH_2), 23.1 (CH_2);

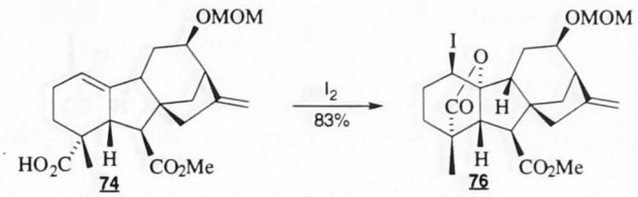
LRMS 390 ($M^{\cdot+}$, 1), 369 (3), 355 (3), 344 (2), 328 (24), 314 (3), 296 (18), 282 (15), 268 (32), 253 (9), 241 (10), 197 (28), 181 (18), 155 (25), 105 (69), 91 (99), 55 (100);

HRMS found 390.2041 ($M^{\cdot+}$), $C_{22}H_{30}O_6$ requires 390.2042.

***ent*-7-Hydroxy-12α-methoxymethoxy-20-norgibberella-1(10),16-diene-19-oic Acid** (**75**, 62 mg, 18%, oil):

^{1}H NMR (300 MHz, $CDCl_3$) 5.41 (1H, m, H1), 4.99 (1H, bs, H17), 4.97 (1H, bs, H'17), 4.72 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.63 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 3.65 (3H, m, H12α, H7,H'7), 3.38 (3H, s, -OCH$_2$O$\underline{CH_3}$), 1.31 (3H, s, H18).

***ent*-10β-Hydroxy-1α-iodo-12α-methoxymethoxy-20-norgibberella-16-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (76)**



2M Aqueous LiOH (321 µl, 0.642 mmol of LiOH) was added to the solution of the ene acid **74** (243 mg, 0.62 mmol) in THF (4.5 ml)/EtOH (9 ml) and the mixture was stirred at room temperature for 30 minutes. A solution of iodine (475 mg, 1.87 mmol) in CH_2Cl_2 (40 ml) was then added and the reaction mixture was stirred overnight. The solution was washed with aqueous $Na_2S_2O_3$ and the inorganic phase was extracted with CH_2Cl_2 (2 x 10 ml). Combined organic layers were dried, concentrated under reduced pressure and the solution chromatographed on silica gel (EtOAc/hexane 1:4) to give the iodide **76** (267 mg, 83%) as an oil:

$[\alpha]_D^{20}$ 39.2° (c 29.4 x 10^{-3}, CH_2Cl_2);

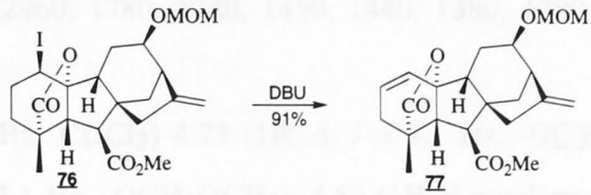
IR ($CDCl_3$) ν_{max} 2940, 1780, 1730, 1660, 1600, 1450, 1440, 1380, 1280, 1200, 1150 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 5.05 (2H, bs, H17, H'17), 4.69 (1H, d, J = 6.7 Hz, -O$\underline{CH_2}$OCH$_3$), 4.63 (1H, d, J = 6.7 Hz, -O$\underline{CH_2}$OCH$_3$), 4.57 (1H, d, $J_{1\alpha,2\alpha}$ = 5.4 Hz, H1α), 3.90 (1H, m, H12α), 3.73 (3H, s, -CO$_2\underline{CH_3}$), 3.40 (1H, d, $J_{6,5}$ = 7.6 Hz, H6), 3.37 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.80 (1H, m, H13), 2.66 (1H, dd, $J_{9,11\alpha}$ = 9.2 Hz, $J_{9,11\beta}$ = 4.8 Hz, H9), 2.61 (1H, d, $J_{5,6}$ = 7.6 Hz, H5), 2.34 (1H, m), 2.18 (3H, m), 1.15 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 178.4 (CO), 173.4 (CO), 146.8 (C16), 110.6 (C17), 94.7 (C10), 94.5 (-O**C**H$_2$OCH$_3$), 75.4 (C12), 55.8 (-OCH$_2$O**C**H$_3$), 55.4 (CH), 52.1 (o), 51.7 (e), 50.3 (o), 49.2 (CH), 49.1 (e), 44.4 (CH), 43.2 (e), 40.3 (e), 31.9 (CH_2), 31.8 (CH_2), 25.7 (C1), 24.1 (CH_2), 16.8 (C18);

LRMS 516 ($M^{\cdot+}$, 0.1), 498 (2), 485 (6), 456 (3), 424 (16), 396 (5), 371 (19), 357 (11), 339 (14), 325 (12), 297 (16), 283 (17), 269 (21), 223 (19), 195 (27), 105 (66), 91 (96), 55 (100).

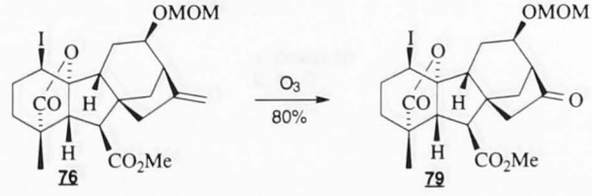
***ent*-10β-Hydroxy-12α-methoxymethoxy-20-norgibberella-1,16-diene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (77)**



DBU (0.5 ml) was added to the solution of iodide **76** (90 mg, 0.174 mmol) in CH_2Cl_2 (5 ml) and the reaction mixture was heated at 40°C for 12 hours. The solution was diluted with CH_2Cl_2 and washed with 1M aqueous HCl. The aqueous phase was extracted with CH_2Cl_2 (3 x 5 ml), the combined organic washings dried and the solvent removed. The residue contained pure product **77** (62 mg, 91%):

^{1}H NMR (300 MHz, $CDCl_3$) 6.20 (1H, dt, $J_{1,2}$ = 9.5 Hz, $J_{1,3\alpha} = J_{1,3\beta}$ = 1.9 Hz, H1), 5.84 (1H, dt, $J_{2,1}$ = 9.5 Hz, $J_{2,3\alpha} = J_{2,3\beta}$ = 3.3 Hz, H2), 5.08 (1H, bs, H17), 5.04 (1H, bs, H'17), 4.70 (1H, d, J = 7.2 Hz, -O$\underline{CH_2}$OCH$_3$), 4.63 (1H, d, J = 7.2 Hz, -O$\underline{CH_2}$OCH$_3$), 4.01 (1H, m, H12α), 3.71 (3H, s, -CO$_2\underline{CH_3}$), 3.37 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.92 (1H, d, $J_{6,5}$ = 8.1 Hz, H6), 2.85 (1H, m, H13), 2.69 (1H, d, $J_{5,6}$ = 8.1 Hz, H5), 1.21 (3H, s, H18).

***ent*-10β-Hydroxy-1α-iodo-12α-methoxymethoxy-16-oxo-17,20-dinorgibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (79)**



Iodide **76** (172 mg, 0.333 mmol) was dissolved in CH_2Cl_2 (17 ml)/pyridine (6 ml) and the solution was cooled to -78°C in an acetone/dry ice bath. A stream of ozonised oxygen was then passed through the solution with efficient stirring until TLC analysis indicated that the reaction was complete (approximately 6 minutes). Me_2S (200 µl) was added, then the reaction mixture was allowed to warm to room temperature and allowed to stand for 30 minutes. The solvent was removed under

reduced pressure and the residue chromatographed on silica gel (EtOAc/hexane 2:3) to afford the 17-norketone **79** (138 mg, 80%) as an oil:

IR ($CDCl_3$) ν_{max} 2960, 1780, 1740, 1450, 1440, 1380, 1280, 1200, 1150, 1100, 1040 cm^{-1};

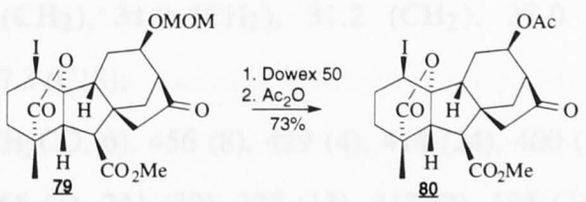
^{1}H NMR (300 MHz, $CDCl_3$) 4.73 (1H, d, J = 7.1 Hz, -OC$\underline{H_2}$OCH$_3$), 4.59 (1H, d overlapped, J = 7.1 Hz, -OC$\underline{H_2}$OCH$_3$), 4.57 (1H, d overlapped, $J_{1\alpha,2\alpha}$ = 5.2 Hz, H1α), 4.23 (1H, m, H12α), 3.74 (3H, s, -CO$_2$C$\underline{H_3}$), 3.48 (1H, d, $J_{6,5}$ = 9.6 Hz, H6), 3.38 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.81 (2H, m, H13, H16), 2.67 (1H, d, $J_{5,6}$ = 9.6 Hz, H5), 2.33 (1H, m), 1.93 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 12.0 Hz, $J_{14\beta,13}$ = 5.3 Hz, H14β), 1.78 (1H, ddd, $J_{11\beta,11\alpha}$ = 14.9 Hz, $J_{11\beta,9}$ = 6.9 Hz, $J_{11\beta,12\alpha}$ = 3.5 Hz, H11β), 1.15 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 214.4 (C16), 178.3 (CO), 172.3 (CO), 95.4 (C10), 94.5 (-OCH$_2$OCH$_3$), 73.3 (C12), 55.9 (-OCH$_2$O**C**H$_3$), 55.2 (CH), 52.6 (o), 52.5 (o), 50.7 (o), 49.9 (e), 49.8 (e), 49.6 (e), 49.5 (CH), 36.3 (CH_2), 31.9 (CH_2), 31.4 (CH_2), 26.6 (C1), 25.6 (CH_2), 17.2 (C18);

LRMS 518 ($M^{\cdot+}$, 0.5), 490 (6), 458 (19), 445 (1), 427 (2), 414 (3), 359 (2), 331 (4), 313 (2), 299 (8), 285 (7), 271 (5), 241 (19), 181 (17), 105 (24), 91 (37), 55 (100);

HRMS found 518.0802 ($M^{\cdot+}$), $C_{21}H_{27}IO_7$ requires 518.0802.

Replacement of the 12-MOM group with the acetyl moiety



79 80

Dowex 50 (1.1 g) was added to the solution of the 12-methoxymethyl derivative **79** (133 mg, 0.257 mmol) in MeOH (22 ml)/H_2O (2.2 ml) and the mixture was heated at 60°C until TLC indicated that the starting material had been replaced by a more polar compound (approximately 72 hours). The mixture was then filtered through a plug of sand and the solvent removed under reduced pressure. The residue contained the pure 12-alcohol (119 mg, 97%):

^{1}H NMR (300 MHz, $CDCl_3$) 4.54 (1H, d, $J_{1\alpha,2\alpha}$ = 5.6 Hz, H1α), 4.37 (1H, m, H12α), 3.74 (3H, s, -$CO_2\underline{CH_3}$), 3.49 (1H, d, $J_{6,5}$ = 11.1 Hz, H6), 2.79 (1H, dd, $J_{9,11\alpha}$ = 12.5 Hz, $J_{9,11\beta}$ = 7.7 Hz, H9), 2.70 (1H, d overlapped, $J_{5,6}$ = 11.1 Hz, H5), 2.69 (1H, m overlapped, H13), 1.15 (3H, s, H18).

This material was dissolved in CH_2Cl_2 (8 ml) and the solution was treated with Et_3N (210 µl, 1.51 mmol) and Ac_2O (140 µl, 1.48 mmol). When TLC analysis revealed that the acetylation was complete, the solution was concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 2:3) to afford **ent-12α-Acetoxy-10β-hydroxy-1α-iodo-16-oxo-17,20-dinorgibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**<u>80</u>**, 97 mg, 75%) as an oil:

IR ($CDCl_3$) ν_{max} 2960, 1780, 1740, 1600, 1450, 1440, 1370, 1250, 1170, 1100, 1040 cm^{-1};

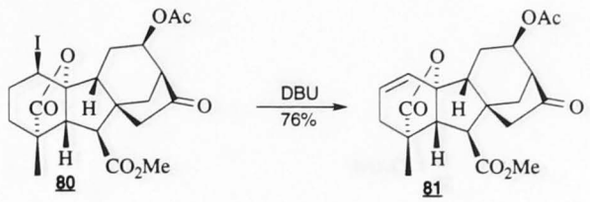
^{1}H NMR (300 MHz, $CDCl_3$) 5.26 (1H, ddd, $J_{12\alpha,13}$ = 8.7 Hz, $J_{12\alpha,11\alpha}$ = 6.4 Hz, $J_{12\alpha,11\beta}$ = 2.2 Hz, H12α), 4.54 (1H, d, $J_{1\alpha,2\alpha}$ = 5.0 Hz, H1α), 3.75 (3H, s, -$CO_2\underline{CH_3}$), 3.50 (1H, d, $J_{6,5}$ = 10.0 Hz, H6), 2.94 (1H, dd, $J_{13,12\alpha}$ = 8.7 Hz, $J_{13,14\beta}$ = 4.3 Hz, H13), 2.77 (1H, dd, $J_{9,11\alpha}$ = 11.4 Hz, $J_{9,11\beta}$ = 6.4 Hz, H9), 2.69 (1H, d, $J_{5,6}$ = 10.0 Hz, H5), 2.33 (1H, m), 2.04 (3H, s, -OCO$\underline{C}H_3$), 1.73 (1H, ddd, $J_{11\beta,11\alpha}$ = 15.2 Hz, $J_{11\beta,9}$ = 6.4 Hz, $J_{11\beta,12\alpha}$ = 2.2 Hz, H11β), 1.14 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 213.8 (C16), 178.1 (CO), 172.1 (CO), 170.6 (CO), 94.2 (C10), 70.5 (C12), 55.1 (CH), 52.7 (o), 50.1 (e), 50.0 (e), 49.7 (e), 49.5 (o), 49.0 (CH), 35.7 (CH_2), 31.9 (CH_2), 31.2 (CH_2), 27.0 (C1), 24.7 (CH_2), 21.3 (-OCOCH_3), 17.3 (C18);

LRMS 474 ($M^{\cdot+}$-CH_2CO, 6), 456 (8), 429 (4), 414 (24), 400 (1), 347 (2), 315 (2), 301 (3), 269 (4), 255 (3), 241 (30), 227 (13), 213 (3), 195 (7), 183 (31), 91 (40), 55 (100);

HRMS found 474.0541 ($M^{\cdot+}$-CH_2CO), $C_{19}H_{23}IO_6$ requires 474.0539.

ent-12α-Acetoxy-10β-hydroxy-16-oxo-17,20-dinorgibberella-1-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (81)



DBU (360 µl, 2.41 mmol) was added to a solution of iodide **80** (93 mg, 0.180 mmol) in CH_2Cl_2 (8 ml) and the reaction mixture was heated at 40°C overnight. The solution was diluted with CH_2Cl_2 and washed with aqueous 1M HCl. The aqueous phase was extracted with CH_2Cl_2 (3 x 5 ml), the combined organic layers dried and the solvent removed. Chromatography of the residue (EtOAc/hexane 2:3) on silica gel afforded olefin **81** (53 mg, 76%) as an oil:

IR ($CDCl_3$) ν_{max} 2960, 1770, 1740, 1600, 1440, 1380, 1250, 1180, 1100, 1030 cm^{-1};

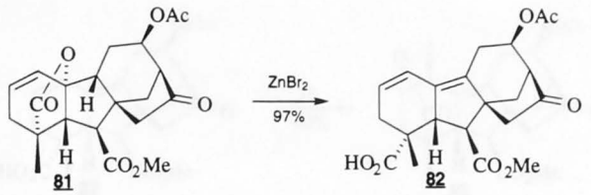
^{1}H NMR (300 MHz, $CDCl_3$) 6.20 (1H, dt, $J_{1,2}$ = 9.3 Hz, $J_{1,3\alpha} = J_{1,3\beta}$ = 2.1 Hz, H1), 5.91 (1H, dt, $J_{2,1}$ = 9.3 Hz, $J_{2,3\alpha} = J_{2,3\beta}$ = 3.6 Hz, H2), 5.30 (1H, m, H12α), 3.75 (3H, s, $-CO_2\underline{CH_3}$), 2.99 (1H, d overlapped, $J_{6,5}$ = 10.7 Hz, H6), 2.98 (1H, m overlapped, H13), 2.79 (1H, d, $J_{5,6}$ = 10.7 Hz, H5), 2.03 (3H, s, $-OCO\underline{CH_3}$), 1.22 (3H, s, H18).

^{13}C NMR (75.5 MHz, $CDCl_3$) 214.0 (C16), 179.2 (CO), 172.4 (CO), 170.5 (CO), 131.7 (C1 or C2), 129.4 (C1 or C2), 90.2 (C10), 70.5 (C12), 56.5 (CH), 52.7 (o), 52.5 (o), 51.1 (C), 50.6 (C), 49.6 (CH), 49.0 (CH), 48.3 (CH_2), 38.2 (CH_2), 35.6 (CH_2), 24.8 (CH_2), 21.3 ($-OCOCH_3$), 17.9 (C18);

LRMS 388 ($M^{\cdot+}$, 0.5), 357 (6), 346 (12), 328 (15), 314 (4), 300 (12), 286 (49), 268 (4), 255 (6), 242 (26), 209 (6), 195 (17), 181 (64), 153 (25), 129 (27), 91 (58), 55 (100);

HRMS found 388.1521 ($M^{\cdot+}$), $C_{21}H_{24}O_7$ requires 388.1522.

***ent*-12α-Acetoxy-16-oxo-17,20-dinorgibberella-1,9-diene-7,19-dioic Acid 7-(Methyl ester) (82)**



$ZnBr_2$ (0.5 g) was added to a stirred solution of olefin **81** (38 mg, 0.098 mmol) in Et_2O and the reaction mixture was stirred while being exposed to atmospheric moisture until the reagent was completely dissolved (approximately 3 hours). The flask was then stoppered and the solution was stirred for a further 12 hours. TLC analysis of the reaction mixture revealed the complete conversion of the starting material into a strongly "UV-active" polar compound showing a streaking spot on the TLC plate. The solution was diluted with EtOAc, washed with ice-cold aqueous 1M HCl, brine and the solvent evaporated *in vacuo*. The residue contained the pure diene acid **82** (37 mg, 97%):

IR ($CDCl_3$) ν_{max} 2960, 1740, 1700, 1470, 1440, 1380, 1240, 1170, 1100, 1040 cm^{-1};

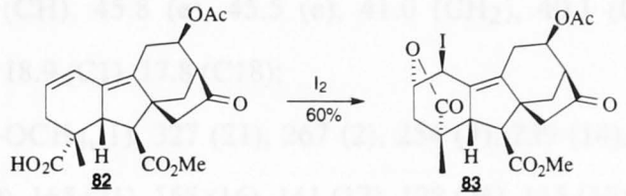
^{1}H NMR (300 MHz, $CDCl_3$) 6.23 (1H, dd, $J_{1,2}$ = 9.8 Hz, J = 2.4 Hz, H1), 5.86 (1H, m, H2), 4.96 (1H, ddd, $J_{12\alpha,11\beta}$ = 10.7 Hz, $J_{12\alpha,11\alpha}$ = 6.8 Hz, $J_{12\alpha,13}$ = 3.7 Hz, H12α), 3.73 (3H, s, -$CO_2\underline{CH_3}$), 3.63 (1H, d, $J_{6,5}$ = 8.3 Hz, H6), 3.41 (1H, dd, $J_{5,6}$ = 8.3 Hz, J = 4.4 Hz, H5), 3.04 (1H, dd, $J_{11\alpha,11\beta}$ = 15.6 Hz, $J_{11\alpha,12\alpha}$ = 6.8 Hz, H11α), 2.74 (2H, m, H13 + 1), 2.32 (1H, dd, J_1 = 12.0 Hz, J_2 = 6.0 Hz), 2.24 (1H, br d, $J_{15\beta,15\alpha}$ = 18.7 Hz, H15β), 2.08 (3H, s overlapped, -OCO$\underline{CH_3}$), 1.90 (1H, dd, $J_{14\alpha,14\beta}$ = 12.0 Hz, $J_{14\alpha,15\beta}$ = 2.8 Hz, H14α), 1.26 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 215.4 (C16), 181.5 (CO), 174.6 (CO), 170.9 (CO), 132.5 (C9 or C10), 130.2 (C9 or C10), 129.9 (C1 or C2), 121.5 (C1 or C2), 72.8 (C12), 56.2 (CH_2), 55.0 (CH), 52.3 (o), 51.7 (o), 49.9 (CH), 46.9 (e), 45.0 (e), 41.4 (e), 38.7 (CH_2), 27.6 (CH_2), 24.4 (C18), 21.5 (-OCOCH_3);

LRMS 328 ($M^{\cdot+}$-AcOH, 13), 279 (3), 254 (28), 240 (15), 211 (5), 195 (6), 181 (100), 165 (17), 155 (10), 149 (60), 128 (10);

HRMS found 328.1311 ($M^{\cdot+}$-AcOH), $C_{19}H_{20}O_5$ requires 328.1311.

***ent*-12α-Acetoxy-2β-hydroxy-1α-iodo-16-oxo-17,20-dinorgibberella-9-ene-7,19-dioic Acid 7-(Methyl ester) 19,2-Lactone (83)**



2M Aqueous LiOH (49 µl, 0.098 mmol of LiOH) was added to a solution of the diene acid **82** (38 mg, 0.098 mmol) in THF (0.7 ml)/EtOH (1.4 ml) and the mixture was stirred at room temperature for 30 minutes. A solution of iodine (75 mg, 0.295 mmol) in CH_2Cl_2 (7 ml) was then added and the reaction mixture was stirred for 30 minutes. The solution was washed with aqueous $Na_2S_2O_3$ and the inorganic phase was extracted with CH_2Cl_2 (2 x 10 ml). The combined organic layers were dried, solvent removed and the residue redissolved in dry CH_2Cl_2 (3 ml). This solution was treated with Et_3N (30 µl, 0.215 mmol) and Ac_2O (20 µl, 0.212 mmol) in order to reacetylate the 12-hydroxy group (the 12-acetoxy function was partially lost during the iodolactonization process). When TLC analysis indicated that the reaction was complete, the solution was concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 2:3) to afford iodolactone **83** (30 mg, 60%) as an oil:

IR ($CDCl_3$) ν_{max} 2960, 1770, 1750, 1740, 1460, 1440, 1380, 1240, 1170, 1040 cm^{-1};

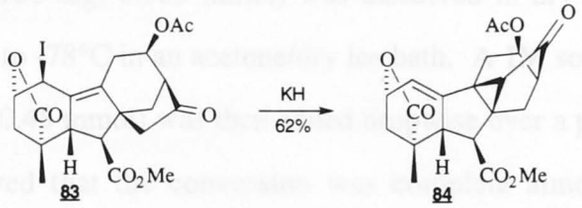
^{1}H NMR (300 MHz, $CDCl_3$) 5.05 (1H, d, $J_{1\alpha,2\beta}$ = 3.4 Hz, H1α), 4.96 (1H, ddd, $J_{12\alpha,11\beta}$ = 10.6 Hz, $J_{12\alpha,11\alpha}$ = 7.0 Hz, $J_{12\alpha,13}$ = 3.7 Hz, H12α), 4.76 (1H, dd, $J_{2\beta,3\alpha}$ = 5.9 Hz, $J_{2\beta,1\alpha}$ = 3.4 Hz, H2β), 3.75 (3H, s, -$CO_2\underline{CH_3}$), 3.62 (1H, dd, $J_{5,6}$ = 8.7 Hz, $J_{5,11\beta}$ = 4.9 Hz, H5), 3.01 (1H, d overlapped, $J_{6,5}$ = 8.7 Hz, H6), 3.00 (1H, dd overlapped, $J_{11\alpha,11\beta}$ = 17.0 Hz, $J_{11\alpha,12\alpha}$ = 7.0 Hz, H11α), 2.83 (1H, d, $J_{3\beta,3\alpha}$ = 12.7 Hz, H3β), 2.74 (1H, m, H13), 2.35 (2H, m, H3α, H14β), 2.08 (3H, s overlapped, -OCO$\underline{CH_3}$), 2.05 (1H, br dd overlapped, $J_{15\beta,15\alpha}$ = 18.0 Hz, $J_{15\beta,14\alpha}$ = 3.3 Hz, H15β), 1.93 (1H, d, $J_{15\alpha,15\beta}$ = 18.0 Hz, H15α), 1.75 (1H, ddd overlapped, $J_{11\beta,11\alpha}$ = 17.0 Hz, $J_{11\beta,12\alpha}$ = 10.6 Hz, $J_{11\beta,5}$ = 4.9 Hz, H11β), 1.68 (1H, dd overlapped, $J_{14\alpha,14\beta}$ = 12.4 Hz, $J_{14\alpha,15\beta}$ = 3.3 Hz, H14α), 1.15 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 213.4 (C16), 178.0 (CO), 172.6 (CO), 170.6 (CO), 143.0 (C9 or C10), 129.6 (C9 or C10), 71.6 (C12), 55.1 (e), 52.7 (o), 52.6 (o), 52.5 (o), 51.0 (CH), 49.8 (CH), 45.8 (e), 45.5 (e), 41.0 (CH_2), 40.1 (CH_2), 27.7 (CH_2), 21.4 (-OCO**C**H_3), 18.9 (C1), 17.8 (C18);

LRMS 483 ($M^{\cdot+}$-OCH_3, 1), 327 (21), 267 (2), 254 (7), 239 (14), 225 (20), 207 (3), 195 (15), 181 (100), 165 (21), 155 (16), 141 (17), 128 (36), 115 (17);

HRMS found 483.0307 ($M^{\cdot+}$-OCH_3), $C_{20}H_{20}IO_6$ requires 483.0305.

***ent*-12α-Acetoxy-2β-hydroxy-16-oxo-17,20-dinor-9α,15α-cyclogibberella-1(10)-ene-7,19-dioic Acid 7-(Methyl ester) 19,2-Lactone (84)**



83 84

Compound **83** (26 mg, 0.051 mmol) was dissolved in dry THF (2.5 ml) under an argon atmosphere and the solution was cooled to 0°C. KH (20 mg, 0.5 mmol) was then added and the resulting suspension was vigorously stirred for 1 hour, whereupon TLC monitoring revealed that the reaction was complete. The reaction mixture was diluted with dry THF (3 ml) and filtered through Celite under argon. The flask was rinsed with dry THF (3 x 3 ml) and the washings filtered through the Celite column. The filtrate was concentrated under reduced pressure and the solution chromatographed on silica gel to give the 9,15-cyclo compound **84** (12 mg, 62%):

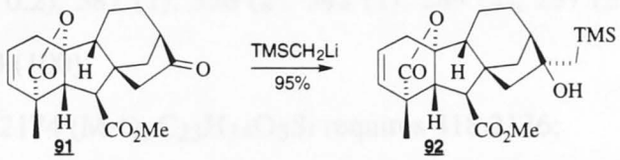
^{1}H NMR (300 MHz, $CDCl_3$) 5.99 (1H, dd, $J_{1,2\beta}$ = 5.3 Hz, $J_{1,5}$ = 2.6 Hz, H1), 5.05 (1H, dt, $J_{12\alpha,11\alpha}$ = 9.1 Hz, $J_{12\alpha,11\beta}$ = $J_{12\alpha,13}$ = 3.6 Hz, H12α), 4.85 (1H, t, $J_{2\beta,1}$ = $J_{2\beta,3\alpha}$ = 5.3 Hz, H2β), 3.77 (3H, s, -$CO_2\underline{CH_3}$), 2.98 (1H, d, $J_{6,5}$ = 9.5 Hz, H6), 2.87 (1H, dd, $J_{5,6}$ = 9.5 Hz, $J_{5,1}$ = 2.6 Hz, H5), 2.58 (1H, dd, $J_{11\alpha,11\beta}$ = 15.2 Hz, $J_{11\alpha,12\alpha}$ = 9.1 Hz, H11α), 2.45 (1H, m, H13), 2.02 (3H, s, -OCO$\underline{CH_3}$), 1.91 (1H, d, $J_{14\alpha,14\beta}$ = 12.5 Hz, H14α), 1.21 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 207.1 (C16), 178.2 (CO), 172.5 (CO), 170.4 (CO), 147.0 (C10), 121.0 (C1), 72.9 (C2 or C12), 71.7 (C2 or C12), 49.1 (-CO_2**C**H_3),

47.2 (CH), 47.0 (e), 46.5 (CH), 45.4 (e), 44.3 (e), 42.8 (e), 37.1 (CH), 28.4 (CH_2), 27.3 (CH_2), 21.3 (-$OCOCH_3$), 20.0 (C18).

5.3.2 3-hydroxy series of compounds

Reaction of model compound 91 with $TMSCH_2Li$



91 92

Ketone **91** (100 mg, 0.303 mmol) was dissolved in dry THF (4 ml) and the solution was cooled to -78°C in an acetone/dry ice bath. A 1M solution of $TMSCH_2Li$ in hexane (0.45 ml, 0.45 mmol) was then added dropwise over a period of 10 minutes. TLC analysis showed that the conversion was complete almost immediately (*ca* 5 minutes). A saturated aqueous solution of NH_4Cl (a few drops) was added to the reaction mixture and the resulting solution was stirred for 5 minutes. The solution was diluted with Et_2O, washed with brine, dried and the solvent removed. Chromatography of the residue on silica gel (EtOAc/hexane 1:3) afforded in order of elution:

16-*epi*-*ent*-16,10β-Dihydroxy-17-trimethylsilyl-20-norgibberella-2-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (**92**, 108 mg, 95%) as an oil:

IR ($CDCl_3$) ν_{max} 3600, 2960, 1770, 1730, 1440, 1380, 1250, 1200, 1160, 990 cm^{-1};

H NMR (300 MHz, $CDCl_3$) 5.79 (1H, dm, $J_{2,3}$ = 9.2 Hz, H2), 5.64 (1H, dm, $J_{3,2}$ = 9.2 Hz, H3), 3.70 (3H, s, -$CO_2\underline{CH_3}$), 2.72 (1H, d, $J_{6,5}$ = 10.4 Hz, H6), 2.58 (1H, ddd overlapped, $J_{1,1'}$ = 18.7 Hz, $J_{1,2}$ = 3.4 Hz, $J_{1,3}$ = 1.7 Hz, H1), 2.57 (1H, d overlapped, $J_{5,6}$ = 10.4 Hz, H5), 2.33 (1H, dt, $J_{1,1'}$ = 18.7 Hz, $J_{1',2}$ = $J_{1',3}$ = 2.7 Hz, H1'), 2.08 (1H, dd overlapped, $J_{9,11\alpha}$ = 10.8 Hz, $J_{9,11\beta}$ = 6.8 Hz, H9), 1.99 (1H, m overlapped, H13), 1.80 (1H, dd overlapped, J_1 = 11.8 Hz, J_2 = 3.0 Hz), 1.55 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 13.1 Hz, $J_{14\beta,13}$ = 3.1 Hz, H14β), 1.42 (1H, d,

$J_{14\alpha,14\beta}$ = 13.1 Hz, H14α), 1.20 (3H, s overlapped, H18), 1.10 (2H, m overlapped, H17), 0.05 (9H, s, -CH_2TMS);

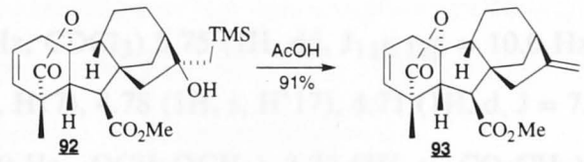
^{13}C NMR (75.5 MHz, $CDCl_3$) 177.9 (CO), 173.0 (CO), 132.1 (C2 or C3), 127.8 (C2 or C3), 92.4 (C10), 80.6 (C16), 54.7 (CH), 54.3 (e), 54.1 (CH), 52.9 (e), 52.3 (-CO_2**C**H_3), 51.8 (CH), 48.0 (e), 46.9 (CH), 37.2 (CH_2), 36.1 (CH_2), 35.3 (CH_2), 20.6 (CH_2), 16.0 (C17), 15.2 (C18), 0.5 (-Si(**C**H_3)$_3$);

LRMS 418 ($M^{\cdot+}$, 0.2), 387 (1), 356 (2), 342 (1), 289 (2), 257 (3), 223 (2), 199 (3), 184 (5), 143 (5), 73 (100);

HRMS found 418.2174 ($M^{\cdot+}$), $C_{23}H_{34}O_5Si$ requires 418.2176;

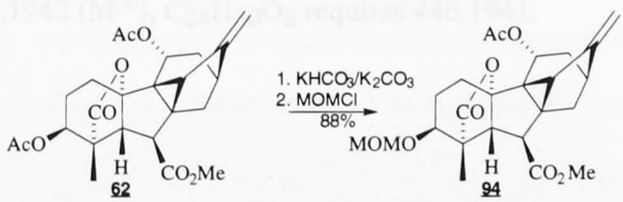
recovered starting material (10 mg).

Decomposition of Peterson adduct 92



Compound **92** (14 mg, 0.033 mmol) was dissolved in neat AcOH (1.5 ml) and the solution was maintained at room temperature. TLC analysis after 7 days showed approximately 70% conversion into olefin **93**. The elimination was then driven to completion by heating the solution at 50°C for 48 hours. The solvent was removed under reduced pressure and the residue chromatographed on silica gel (hexane/EtOAc 9:1) to afford olefin **93**[63] (10 mg, 91%).

***ent*-11β-Acetoxy-10β-hydroxy-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberella-16-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (94)**



Diacetate **62** (500 mg, 1.13 mmol) was dissolved in methanol (50 ml) and an aqueous solution of $KHCO_3/K_2CO_3$ was added dropwise (1.3 ml, 0.5 M, 50 mg $KHCO_3$/70 mg K_2CO_3 in 1 ml of solution). The mixture was stirred at room temperature for two hours at which stage TLC revealed that the reaction was complete. The solution was then poured into brine and the resultant mixture extracted with EtOAc. The organic layer was dried, solvent evaporated *in vacuo* and the crude alcohol redissolved in CH_2Cl_2 (50 ml) and DIPEA (12 ml). MOMCl (5 ml, 65.8 mmol) was added to the solution followed by DMAP (50 mg, 0.409 mmol). TLC analysis indicated that the starting material was consumed in 48 hours. The solution was then diluted with EtOAc, subjected to standard work-up, dried and the solvent evaporated under reduced pressure. Chromatography of the residue (EtOAc/hexane 3:7) afforded the desired 3-methoxymethoxy compound **94** (444 mg, 88%) as an oil:

IR ($CDCl_3$) ν_{max} 2960, 1770, 1730, 1670, 1600, 1440, 1370, 1250, 1190, 1170, 1150 cm^{-1};

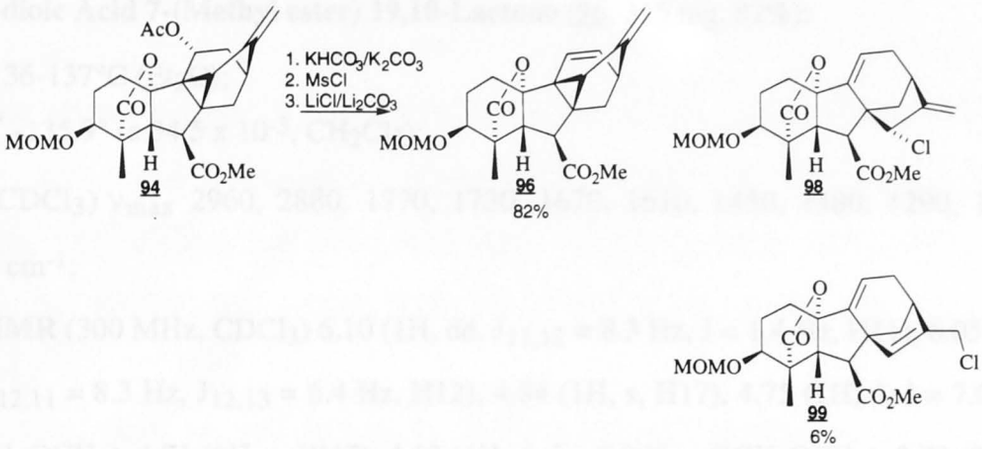
^{1}H NMR (300 MHz, $CDCl_3$) 5.75 (1H, dd, $J_{11\beta,12\beta}$ = 10.0 Hz, $J_{11\beta,12\alpha}$ = 3.3 Hz, H11β), 4.80 (1H, s, H17), 4.78 (1H, s, H'17), 4.71 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.60 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 3.72 (3H, s, -CO$_2\underline{CH_3}$), 3.59 (1H, m, H3α), 3.38 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.98 (1H, d, $J_{6,5}$ = 9.4 Hz, H6), 2.71 (1H, d, $J_{5,6}$ = 9.4 Hz, H5), 2.45 (1H, m, H13), 2.24 (1H, m), 2.10 (3H, s, OCO$\underline{CH_3}$), 1.13 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 176.6 (CO), 172.1 (CO), 170.7 (CO), 149.9 (C16), 103.0 (C17), 95.5 (-O**C**H$_2$OCH$_3$), 92.9 (C10), 75.1 (C3), 64.5 (C11), 55.5 (-OCH$_2$O**C**H$_3$), 51.8 (-CO$_2$**C**H$_3$), 51.5 (C), 49.4 (CH), 45.4 (CH) 41.9 (C), 41.0 (C), 37.2 (CH), 36.9 (CH_2), 32.0 (CH_2), 29.8 (C15), 24.4 (CH_2), 24.1 (CH_2), 21.5 (-OCO**C**H$_3$), 13.7 (C18);

LRMS 446 ($M^{\cdot+}$, 34), 386 (60), 356 (8), 342 (24), 324 (25), 296 (34), 280 (100), 253 (29), 237 (27), 221 (94), 199 (49), 180 (32), 165 (27), 91 (18);

HRMS found 446.1942 ($M^{\cdot+}$), $C_{24}H_{30}O_8$ requires 446.1941.

Preparation of diene 96



Acetate **94** (444 mg, 0.996 mmol) was dissolved in MeOH (50 ml). Aqueous $KHCO_3/K_2CO_3$ (5 ml, 0.5 M, 50 mg $KHCO_3$/70 mg K_2CO_3 in 1 ml of solution) was added to the solution and the mixture was stirred at room temperature for 12 hours. K_2CO_3 (140 mg, 1.01 mmol) was then added and stirring was continued until TLC monitoring indicated that the reaction was complete (*ca* 36 hours). The solution was poured into brine, the resulting mixture extracted with EtOAc, the organic layer dried and the solvent evaporated under reduced pressure.

The crude alcohol was redissolved in dry CH_2Cl_2 (30 ml) and Et_3N (1.8 ml, 12.91 mmol). After cooling in an ice bath, methanesulfonyl chloride (0.7 ml, 9.04 mmol) was added followed by DMAP (50 mg, 0.409 mmol) and the reaction mixture was allowed to warm to room temperature. TLC analysis after 12 hours revealed that the reaction was complete. The solution was diluted with EtOAc, subjected to standard work-up, dried and the solvent evaporated *in vacuo*; the last traces of solvent and the excess of the reagent were removed under high vacuum at 50°C. The residue was redissolved in dry DMF (30 ml); dry LiCl (1.3 g) and dry Li_2CO_3 (1.4 g) were added and the reaction mixture was stirred at 80°C for 5 hours. The solvent was removed under high vacuum and the solid material was redissolved in EtOAc/brine. The organic phase was dried, concentrated under reduced pressure and the solution chromatographed on silica gel (EtOAc/hexane 1:3) to afford in order of elution:

***ent*-10β-Hydroxy-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberella-11,16-diene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**96**, 315 mg, 82%):

mp 136-137°C (Et_2O);

$[\alpha]_D^{20}$ -135.9° (c 34.5 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2960, 2880, 1770, 1730, 1670, 1610, 1450, 1380, 1290, 1150, 1110 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 6.10 (1H, dd, $J_{11,12}$ = 8.3 Hz, J = 1.4 Hz, H11), 6.05 (1H, dd, $J_{12,11}$ = 8.3 Hz, $J_{12,13}$ = 6.4 Hz, H12), 4.84 (1H, s, H17), 4.75 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.71 (1H, s, H'17), 4.63 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 3.73 (3H, s, -$CO_2\underline{CH_3}$), 3.69 (1H, m, H3α), 3.40 (3H, s, -$OCH_2O\underline{CH_3}$), 2.98 (1H, d overlapped, $J_{6,5}$ = 9.3 Hz, H6), 2.97 (1H, m overlapped, H13), 2.87 (1H, d, $J_{5,6}$ = 9.3 Hz, H5), 2.19 (1H, s overlapped, H15), 2.18 (1H, m overlapped), 1.86 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 10.8 Hz, $J_{14\beta,13}$ = 5.1 Hz, H14β), 1.18 (3H, s, H18), 0.98 (1H, d, $J_{14\alpha,14\beta}$ = 10.8 Hz, H14α);

^{13}C NMR (75.5 MHz, $CDCl_3$) 177.7 (CO), 173.2 (CO), 148.6 (C16), 129.0 (C11), 121.4 (C12), 103.5 (C17), 96.2 (-O**C**H$_2$OCH$_3$), 92.7 (C10), 75.9 (C3), 56.2 (-OCH$_2$O**C**H$_3$), 53.6 (C), 52.5 (-CO_2**C**H$_3$), 50.5 (CH), 45.3 (CH), 42.5 (C), 41.9 (CH), 41.5 (C), 30.5 (CH_2), 28.0 (C15), 26.4 (CH_2), 25.3 (CH_2), 14.4 (C18);

LRMS 386 ($M^{\cdot+}$, 24), 354 (2), 324 (4), 310 (10), 296 (2), 280 (20), 265 (13), 237 (9), 221 (100), 181 (21), 165 (25), 115 (32);

HRMS found 386.1730 ($M^{\cdot+}$), $C_{22}H_{26}O_6$ requires 386.1729.

Anal. Found: C, 68.13; H, 6.49. Calcd for $C_{22}H_{26}O_6$: C, 68.38; H, 6.78.

***ent*-17-Chloro-10β-hydroxy-3α-methoxymethoxy-20-norgibberella-9(11),15-diene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**99**) and ***ent*-15β-Chloro-10β-hydroxy-3α-methoxymethoxy-20-norgibberella-9(11),16-diene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**98**) (27 mg of the mixture of both compounds, 6% overall yield):

99: **^{1}H NMR** (300 MHz, $CDCl_3$) 6.10 (1H, s, H15), 5.71 (1H, m, H11), 4.76 (1H, d overlapped, J = 7.1 Hz, -O$\underline{CH_2}$OCH$_3$), 4.66 (1H, d overlapped, J = 7.1 Hz,

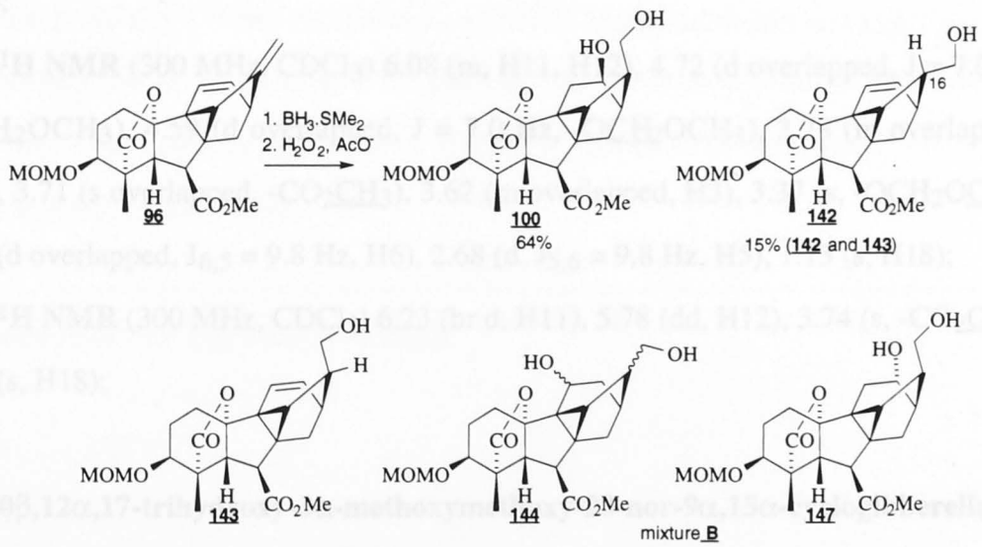
-O$\underline{CH_2}$OCH$_3$), 3.71 (3H, s, -CO$_2\underline{CH_3}$), 3.40 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.23 (1H, d, $J_{5,6}$ = 9.9 Hz, H5), 2.98 (1H, m, H13), 2.62 (1H, d, $J_{6,5}$ = 9.9 Hz, H6), 1.17 (3H, s, H18);

$\underline{98}$: **^{1}H NMR** (300 MHz, $CDCl_3$) 5.98 (1H, m, H11), 5.31 (1H, s, H17), 5.27 (1H, s, H'17), 4.77 (1H, d overlapped, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.68 (1H, d overlapped, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 3.69 (3H, s, -CO$_2\underline{CH_3}$), 3.42 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.32 (1H, d, $J_{5,6}$ = 10.4 Hz, H5), 3.02 (1H, m, H13), 2.70 (1H, d, $J_{6,5}$ = 10.4 Hz, H6), 1.26 (3H, s, H18);

LRMS 422 ($M^{\cdot+}$, 14), 387 ($M^{\cdot+}$-Cl, 9), 360 (10), 332 (4), 316 (60), 281 (34), 221 (46), 195 (20), 181 (27), 165 (38), 115 (55), 55 (100) (**mixture**) ;

HRMS found 422.1496 ($M^{\cdot+}$), $C_{22}H_{27}O_6Cl^{35}$ requires 422.1496.

Hydroboration of diene $\underline{96}$



Diene **$\underline{96}$** (315 mg, 0.815 mmol) was dissolved in dry THF (66 ml) under Ar. The solution was cooled to 0°C and treated with $BH_3.SMe_2$ (4.6 ml, approximately 1.18 mmol; 0.5 ml of 10.2 M $BH_3.SMe_2$ diluted with 20 ml of dry THF), dropwise over a period of 10 minutes. The reaction mixture was allowed to warm to room temperature over 3 hours at which stage TLC indicated that the starting material had almost disappeared. The excess of the reagent was destroyed by the addition of EtOH (3.5 ml) and, after stirring for 5 minutes, the solution was subjected to oxidative work-up: saturated aqueous NaOAc (3.5 ml) was added followed by 30% H_2O_2 (3 ml)

and the resulting mixture was stirred for 48 hours. K_2CO_3 (150 mg, 1.09 mmol) was added after this period and stirring was continued for another 24 hours. The solution was concentrated under reduced pressure, diluted with EtOAc and thoroughly washed with brine. The organic phase was dried, concentrated and chromatographed on silica gel (EtOAc/hexane 1:1 then EtOAc/MeOH 98:2) to afford in order of elution (given yields are based on the amount of diene **96** which reacted with the borane reagent):

recovered starting material (42 mg)

16-*epi*-*ent*-10β,17-Dihydroxy-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberella-11-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (**142**) and ***ent*-10β,17-Dihydroxy-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberella-11-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**143**) (major components of mixture **A**, 43 mg, 15%):

142: **^{1}H NMR** (300 MHz, $CDCl_3$) 6.08 (m, H11, H12), 4.72 (d overlapped, J = 7.0 Hz, $-OCH_2OCH_3$), 4.59 (d overlapped, J = 7.0 Hz, $-OCH_2OCH_3$), 3.78 (m overlapped, H17), 3.71 (s overlapped, $-CO_2CH_3$), 3.62 (m overlapped, H3), 3.37 (s, $-OCH_2OCH_3$), 2.85 (d overlapped, $J_{6,5}$ = 9.8 Hz, H6), 2.68 (d, $J_{5,6}$ = 9.8 Hz, H5), 1.13 (s, H18);

143: **^{1}H NMR** (300 MHz, $CDCl_3$) 6.23 (br d, H11), 5.78 (dd, H12), 3.74 (s, $-CO_2CH_3$), 1.20 (s, H18);

***ent*-10β,12α,17-trihydroxy-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-Methyl Ester 19,10-Lactone** (**100**) as a colourless oil (192 mg, 64%):

$[\alpha]_D^{20}$ +34.0° (c 24.4 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 3610, 3480, 2940, 1770, 1730, 1600, 1450, 1440, 1270, 1150, 1030 cm^{-1};

^{1}H NMR (500 MHz, $CDCl_3$) 4.69 (1H, d, J = 7.1 Hz, $-OCH_2OCH_3$), 4.58 (1H, d, J = 7.1 Hz, $-OCH_2OCH_3$), 3.94 (1H, m, H12α), 3.84 (2H, m, H17), 3.70 (3H, s, $-CO_2CH_3$), 3.60 (1H, m, H3α), 3.35 (3H, s, $-OCH_2OCH_3$), 2.79 (1H, d, $J_{6,5}$ = 9.2 Hz,

H6), 2.72 (1H, d, $J_{5,6}$ = 9.2 Hz, H5), 2.62 (1H, dd, $J_{11\alpha,11\beta}$ = 14.5 Hz, $J_{11\alpha,12\alpha}$ = 10.4 Hz, H11α), 2.34 (1H, m, H16), 2.10 (1H, m, H13), 1.87 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 12.1 Hz, $J_{14\beta,13}$ = 6.2 Hz, H14β), 1.68 (1H, dd, $J_{1\beta,1\alpha}$ = 12.1 Hz, $J_{1\beta,2\alpha}$ = 5.1 Hz, H1β), 1.60 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.1 Hz, H14α), 1.40 (1H, d, J = 3.3 Hz, H15), 1.11 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 177.7 (CO), 173.1 (CO), 95.9 (-O**C**H_2OCH_3), 94.1 (C10), 75.7 (C3), 68.9 (C12), 62.7 (C17), 55.8 (-OCH_2O**C**H_3), 53.3 (C4), 52.0 (-CO_2**C**H_3), 50.2 (C5), 45.2 (C6), 42.6 (C16), 38.5 (C13), 37.9 (C8), 32.1 (C14), 32.0 (C9), 27.5 (C11), 24.8 (C2), 24.4 (C1), 23.0 (C15), 14.1 (C18);

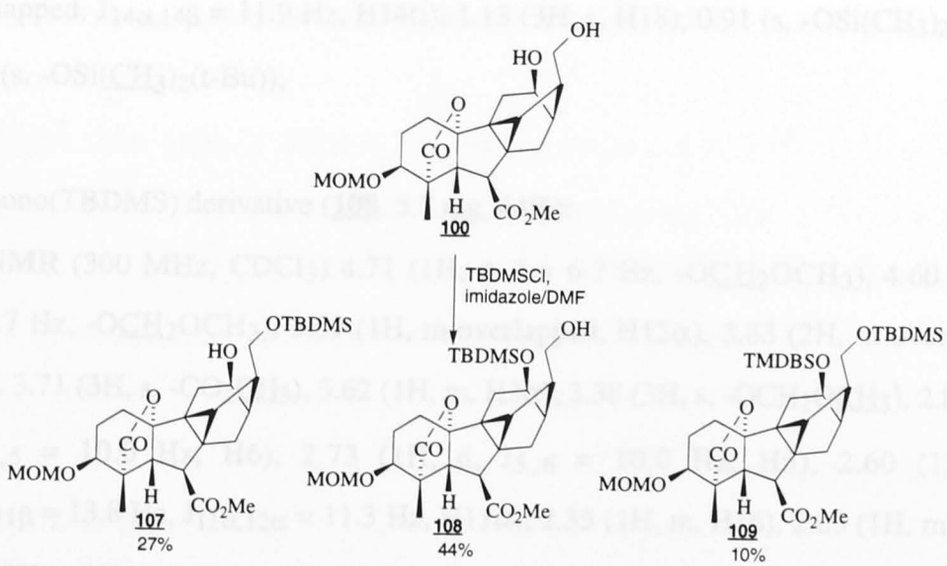
LRMS 404 ($M^{\cdot+}$-H_2O, 32), 376 (10), 359 (4), 342 (15), 328 (6), 316 (14), 298 (58), 268 (13), 241 (19), 225 (30), 181 (38), 149 (49), 91 (51), 55 (100);

HRMS found 404.1835 ($M^{\cdot+}$-H_2O), $C_{22}H_{28}O_7$ requires 404.1835.

mixture **B** (48 mg, 2 major components, indirectly identified as **144** and **147**):

^{1}H NMR (500 MHz, $CDCl_3$) 4.71 and 4.60 (AB systems of two MOM-functions), 3.99 (m), 3.73 (s, -CO_2CH_3), 3.38 (s, -OCH_2OCH_3), 2.86 (d overlapped, J = 9.5 Hz), 2.69 (d, J = 9.5 Hz), 1.14 (s).

Protection of diol 100 with TBDMSCl



Imidazole (3 mg, 0.044 mmol) and TBDMSCl (7 mg, 0.046 mmol) were added to a solution of diol **100** (16 mg, 0.038 mmol) in dry DMF (0.6 ml). TLC analysis after

12 hours showed the presence of three products together with unreacted starting material. The reaction mixture was directly chromatographed on silica gel (EtOAc/hexane 1:1 then EtOAc/MeOH 98:2) to afford in order of elution:

12,17-bis(TBDMS) derivative (**109**, 1.5 mg, 10%):

^{1}H NMR (300 MHz, $CDCl_3$) 4.73 (1H, d, J = 6.6 Hz, -O$\underline{CH_2}$OCH$_3$), 4.62 (1H, d, J = 6.6 Hz, -O$\underline{CH_2}$OCH$_3$), 3.88 (2H, m, H17), 3.78 (1H, m, H12α), 3.70 (3H, s, -CO$_2\underline{CH_3}$), 3.62 (1H, m, H3α), 3.38 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.80 (1H, d, $J_{6,5}$ = 10.1 Hz, H6), 2.76 (1H, d, $J_{5,6}$ = 10.1 Hz, H5), 2.55 (1H, dd, $J_{11\alpha,11\beta}$ = 13.7 Hz, $J_{11\alpha,12\alpha}$ = 10.7 Hz, H11α), 2.27 (1H, m, H16), 1.13 (3H, s, H18), 0.91 (s, -OSi(CH$_3$)$_2$(t-$\underline{Bu}$), 0.85 (s, -OSi(CH$_3$)$_2$(t-$\underline{Bu}$)), 0.07 (s, -OSi($\underline{CH_3}$)$_2$(t-Bu)), 0.06 (s, -OSi($\underline{CH_3}$)$_2$(t-Bu)), 0.00 (s, -OSi($\underline{CH_3}$)$_2$(t-Bu));

17-mono(TBDMS) derivative (**107**, 3.5 mg, 27%):

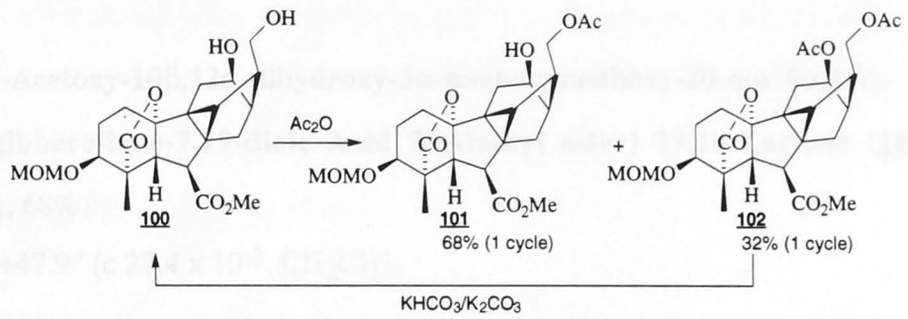
^{1}H NMR (300 MHz, $CDCl_3$) 4.71 (1H, d, J = 6.7 Hz, -O$\underline{CH_2}$OCH$_3$), 4.60 (1H, d, J = 6.7 Hz, -O$\underline{CH_2}$OCH$_3$), 3.95 (2H, m, H17), 3.80 (1H, m, H12α), 3.71 (3H, s, -CO$_2\underline{CH_3}$), 3.62 (1H, m, H3α), 3.38 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.81 (1H, d, $J_{6,5}$ = 9.4 Hz, H6), 2.72 (1H, d, $J_{5,6}$ = 9.4 Hz, H5), 2.65 (1H, dd, $J_{11\alpha,11\beta}$ = 13.8 Hz, $J_{11\alpha,12\alpha}$ = 11.3 Hz, H11α), 2.26 (1H, m, H16), 2.07 (1H, m, H13), 1.63 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 11.9 Hz, H14α), 1.13 (3H, s, H18), 0.91 (s, -OSi(CH$_3$)$_2$(t-$\underline{Bu}$)), 0.10 (s, -OSi($\underline{CH_3}$)$_2$(t-Bu));

12-mono(TBDMS) derivative (**108**, 5.7 mg, 44%):

^{1}H NMR (300 MHz, $CDCl_3$) 4.71 (1H, d, J = 6.7 Hz, -O$\underline{CH_2}$OCH$_3$), 4.60 (1H, d, J = 6.7 Hz, -O$\underline{CH_2}$OCH$_3$), 3.89 (1H, m overlapped, H12α), 3.83 (2H, m overlapped, H17), 3.71 (3H, s, -CO$_2\underline{CH_3}$), 3.62 (1H, m, H3α), 3.38 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.80 (1H, d, $J_{6,5}$ = 10.0 Hz, H6), 2.73 (1H, d, $J_{5,6}$ = 10.0 Hz, H5), 2.60 (1H, dd, $J_{11\alpha,11\beta}$ = 13.8 Hz, $J_{11\alpha,12\alpha}$ = 11.3 Hz, H11α), 2.35 (1H, m, H16), 2.03 (1H, m, H13), 1.14 (3H, s, H18), 0.89 (s, -OSi(CH$_3$)$_2$(t-$\underline{Bu}$)), 0.08 (s, -OSi($\underline{CH_3}$)$_2$(t-Bu));

recovered starting material (6 mg).

Acetylation of diol 100



Compound **100** (192 mg, 0.455 mmol) was dissolved in dry CH_2Cl_2 (20 ml). Et_3N (350 µl, 2.51 mmol) was added to the solution followed by Ac_2O (175 µl, 1.85 mmol). The reaction mixture was maintained at room temperature for 48 hours at which stage TLC indicated that most of the starting material had been consumed and then at 0°C for 72 hours (diol **100** was hardly detectable by TLC at this stage). The solution was concentrated under reduced pressure and chromatographed on SiO_2 (EtOAc/hexane 1:1 followed by 7:3 then neat EtOAc) to afford in order of elution (given yields are based on the amount of diol **100** which reacted with acetic anhydride):

***ent*-12α,17-Diacetoxy-10β-hydroxy-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**102**, oil, 67 mg, 32%):

^{1}H NMR (300 MHz, $CDCl_3$) 4.82 (1H, m, H12α), 4.71 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.61 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.32 (2H, d, $J_{17,16}$ = 8.3 Hz, H17), 3.71 (3H, s, -CO$_2\underline{CH_3}$), 3.64 (1H, m, H3α), 3.39 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.82 (1H, d, $J_{6,5}$ = 9.6 Hz, H6), 2.73 (1H, d overlapped, $J_{5,6}$ = 9.6 Hz, H5), 2.72 (1H, dd overlapped, $J_{11\alpha,11\beta}$ = 14.1 Hz, $J_{11\alpha,12\alpha}$ = 10.8 Hz, H11α), 2.40 (1H, m, H16), 2.21 (1H, m, H13), 2.09 (3H, s, OCO$\underline{CH_3}$), 1.98 (3H, s overlapped, OCO$\underline{CH_3}$), 1.14 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 177.6 (CO), 173.3 (CO), 171.4 (CO), 170.5 (CO) 96.1 (-OCH$_2$OCH$_3$), 93.9 (C10), 75.8 (C3), 71.7 (C12), 65.0 (C17),

56.2 ($-OCH_2OCH_3$), 53.6 (C4), 52.5 ($-CO_2CH_3$), 50.5 (C5), 45.4 (C6), 40.2 (C16), 38.3 (C8), 36.5 (C13), 32.2 (C14), 32.0 (C9), 25.1 (C11), 24.8 (C2), 24.4 (C1), 23.2 (C15), 21.6 ($-OCOCH_3$), 21.4 ($-OCOCH_3$), 14.5 (C18);

***ent*-17-Acetoxy-10β,12α-dihydroxy-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**101**, foam, 133mg, 68%):

$[\alpha]_D^{20}$ +47.9° (c 27.4 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 3600, 2950, 1760, 1730, 1450, 1440, 1370, 1260, 1150, 1040 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 4.72 (1H, d, J = 6.9 Hz, $-OCH_2OCH_3$), 4.61 (1H, d, J = 6.9 Hz, $-OCH_2OCH_3$), 4.42 (2H, d, $J_{17,16}$ = 7.5 Hz, H17), 3.96 (1H, m, H12α), 3.72 (3H, s, $-CO_2CH_3$), 3.63 (1H, m, H3α), 3.38 (3H, s, $-OCH_2OCH_3$), 2.81 (1H, d, $J_{6,5}$ = 9.2 Hz, H6), 2.74 (1H, d, $J_{5,6}$ = 9.2 Hz, H5), 2.64 (1H, dd, $J_{11\alpha,11\beta}$ = 14.4 Hz, $J_{11\alpha,12\alpha}$ = 10.3 Hz, H11α), 2.38 (1H, m, H16), 2.08 (3H, s, $OCOCH_3$), 1.61 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.0 Hz, H14α), 1.14 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 177.5 (CO), 173.1 (CO), 171.1 (CO), 95.8 ($-OCH_2OCH_3$), 93.9 (C10), 75.6 (C3), 69.3 (C12), 65.1 (C17), 55.9 ($-OCH_2OCH_3$), 53.3 (C4), 52.1 ($-CO_2CH_3$), 50.2 (C5), 45.1 (C6), 40.3 (C16), 39.8 (C13), 38.0 (C8), 32.2 (C14), 31.9 (C9), 26.8 (C11), 24.8 (C2), 24.5 (C1), 23.2 (C15), 21.1 ($-OCOCH_3$), 14.2 (C18);

LRMS 462 ($M^{\cdot+}$-2H, 16), 447 (6), 434 (7), 416 (9), 404 ($M^{\cdot+}$-AcOH, 32), 386 (34), 372 (24), 359 (20), 340 (46), 328 (100), 298 (88), 268 (42), 254 (70), 225 (61), 195 (62);

HRMS found 404.1835 ($M^{\cdot+}$-AcOH), $C_{22}H_{28}O_7$ requires 404.1835.

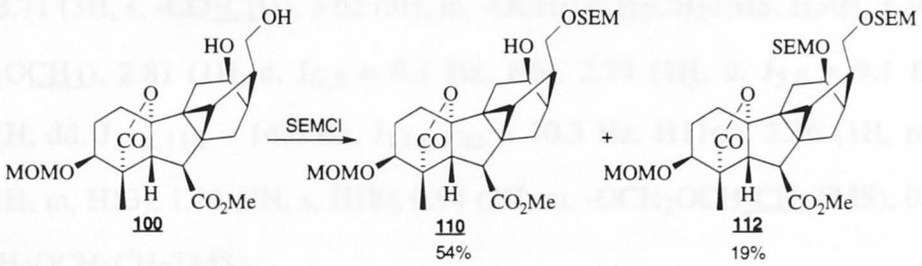
recovered diol **100** (9 mg).

Diacetate **102** was dissolved in MeOH (5 ml) and treated with an aqueous solution of $KHCO_3/K_2CO_3$ (0.4 ml, 0.5 M, 50 mg $KHCO_3$/70 mg K_2CO_3 in 1 ml of solution). When TLC monitoring indicated that the hydrolysis was complete (*ca* 2 hours), the solution was poured into brine, the resulting mixture extracted with

EtOAc, the organic phase dried and the solvent evaporated *in vacuo*. The crude diol **100** (53 mg, identical with the starting compound) together with the recovered material was resubjected to the same acetylation conditions.

After three cycles, monoacetate **101** was obtained in a total quantity of 158 mg (75% overall yield).

Protection of diol 100 with SEMCl



DIPEA (6 µl, 0.034 mmol) and SEMCl (4.2 µl, 0.024 mmol) were added to a solution of diol **100** (10 mg, 0.024 mmol) in dry CH_2Cl_2 at 0°C and the solution was allowed to warm to room temperature over a period of three hours. TLC analysis after 48 hours revealed that a substantial amount of the starting material remained unreacted in the reaction mixture. Pyridine (2 µl, 0.025 mmol) and SEMCl (4.2 µl, 0.024 mmol) were added and the reaction was allowed to proceed for a further 14 hours. Further equivalents of DIPEA (6 µl, 0.034 mmol) and SEMCl (4.2 µl, 0.024 mmol) were then added and, after the period of 6 hours, the solution was concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 1:1) to afford in order of elution:

12,17-bis(SEM) derivative (**112**, 3 mg, 19%):

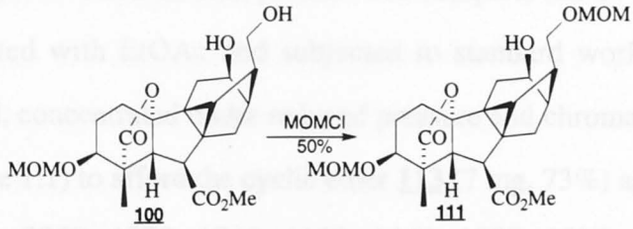
^{1}H NMR (300 MHz, $CDCl_3$) 4.62 (6H, m, 12-O$\underline{CH_2}$OCH$_2$CH$_2$TMS, 17-O$\underline{CH_2}$OCH$_2$CH$_2$TMS, -O$\underline{CH_2}$OCH$_3$), 3.79 (3H, m, H17, H12α), 3.70 (3H, s, -CO$_2\underline{CH_3}$), 3.59 (5H, m, 12-OCH$_2$O$\underline{CH_2}$CH$_2$TMS, 17-OCH$_2$O$\underline{CH_2}$CH$_2$TMS, H3α), 3.37 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.81 (1H, d, $J_{6,5}$ = 9.2 Hz, H6), 2.75 (1H, d, $J_{5,6}$ = 9.2 Hz, H5), 2.59 (1H, dd, $J_{11\alpha,11\beta}$ = 14.3 Hz, $J_{11\alpha,12\alpha}$ = 10.5 Hz, H11α), 2.32 (1H, m, H16), 2.10 (1H, m, H13), 1.59 (1H, d, $J_{14\alpha,14\beta}$ = 12.02 Hz, H14α), 1.12 (3H, s, H18),

0.91 (4H, m, 12-$OCH_2OCH_2\underline{CH_2}TMS$, 17-$OCH_2OCH_2\underline{CH_2}TMS$), 0.00 (6H, s, 12-$OCH_2OCH_2CH_2\underline{TMS}$, 17-$OCH_2OCH_2CH_2\underline{TMS}$);

17-mono(SEM) derivative (**<u>110</u>**, 7 mg, 54%):

^{1}H NMR (300 MHz, $CDCl_3$) 4.71 (2H, s overlapped, -$O\underline{CH_2}OCH_2CH_2TMS$), 4.70 (1H, d overlapped, J = 6.7 Hz, -$O\underline{CH_2}OCH_3$), 4.60 (1H, d, J = 6.7 Hz, -$O\underline{CH_2}OCH_3$), 3.88 (1H, m overlapped, H12α), 3.88 (2H, d overlapped, J = 6.8 Hz, H17), 3.71 (3H, s, -$CO_2\underline{CH_3}$), 3.62 (3H, m, -$OCH_2O\underline{CH_2}CH_2TMS$, H3α), 3.38 (3H, s, -$OCH_2O\underline{CH_3}$), 2.81 (1H, d, $J_{6,5}$ = 9.1 Hz, H6), 2.74 (1H, d, $J_{5,6}$ = 9.1 Hz, H5), 2.65 (1H, dd, $J_{11\alpha,11\beta}$ = 14.0 Hz, $J_{11\alpha,12\alpha}$ = 10.3 Hz, H11α), 2.36 (1H, m, H16), 2.05 (1H, m, H13), 1.13 (3H, s, H18), 0.94 (2H, m, -$OCH_2OCH_2\underline{CH_2}TMS$), 0.02 (3H, s, -$OCH_2OCH_2CH_2\underline{TMS}$).

Protection of diol <u>100</u> with MOMCl

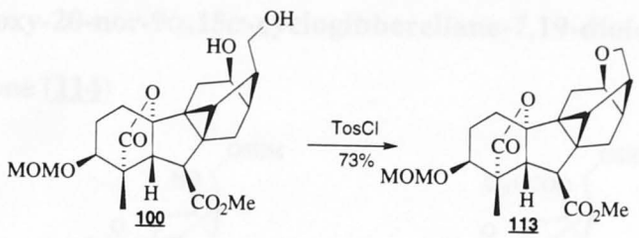


DIPEA (10 µl, 0.057 mmol) and MOMCl (2.5 µl, 0.033 mmol) were added to a solution of compound **<u>100</u>** (11 mg, 0.026 mmol) in dry CH_2Cl_2 (2 ml) at 0°C and the reaction mixture was allowed to warm to room temperature over the period of 1 hour. After 60 hours at room temperature, the solution was concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 4:1) to afford the 17-monoprotected derivative **<u>111</u>** (6 mg, 50%) as an oil:

^{1}H NMR (300 MHz, $CDCl_3$) 4.71 (1H, d, J = 7.0 Hz, 3-$O\underline{CH_2}OCH_3$), 4.67 (2H, s, 17-$O\underline{CH_2}OCH_3$), 4.60 (1H, d, J = 7.0 Hz, 3-$O\underline{CH_2}OCH_3$), 3.88 (3H, m, H17, H12α), 3.71 (3H, s, -$CO_2\underline{CH_3}$), 3.62 (1H, m, H3α), 3.38 (3H, s, -$OCH_2O\underline{CH_3}$), 3.37 (3H, s, -$OCH_2O\underline{CH_3}$), 2.82 (1H, d, $J_{6,5}$ = 8.7 Hz, H6), 2.74 (1H, d, $J_{5,6}$ = 8.7 Hz, H5), 2.65 (1H, dd, $J_{11\alpha,11\beta}$ = 14.6 Hz, $J_{11\alpha,12\alpha}$ = 10.5 Hz, H11α), 2.38 (1H, m, H16),

2.05 (1H, m, H13), 1.87 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 11.6 Hz, $J_{14\beta,13}$ = 5.8 Hz, H14β), 1.62 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 11.6 Hz, H14α), 1.13 (3H, s, H18).

***ent*-12α,17-Epoxy-10β-hydroxy-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>113</u>)**



Toluenesulfonyl chloride (6 mg, 0.031 mmol) and Et_3N (10 µl, 0.072 mmol) were added to a solution of compound **<u>100</u>** (10 mg, 0.024 mmol) in CH_2Cl_2 (1 ml). TLC monitoring after 24 hours showed a very slow conversion into a less polar product. More reagent (12 mg, 0.063 mmol) was added together with a few crystals of DMAP. When TLC revealed that the process was complete (*ca* 24 hours), the reaction mixture was diluted with EtOAc and subjected to standard work-up. The organic solution was dried, concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 1:1) to afford the cyclic ether **<u>113</u>** (7 mg, 73%) as an oil:

IR ($CDCl_3$) ν_{max} 2960, 1770, 1740, 1460, 1440, 1380, 1280, 1200, 1160, 1100, 1040 cm^{-1};

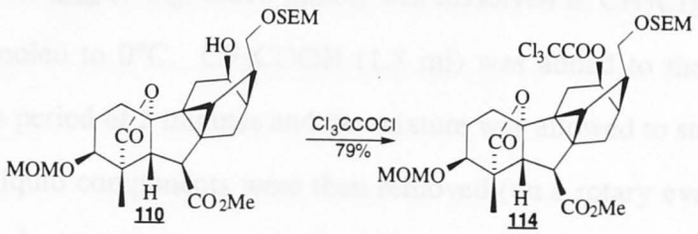
^{1}H NMR (300 MHz, $CDCl_3$) 4.72 (1H, d, J = 7.6 Hz, -O<u>C</u>H_2OCH$_3$), 4.61 (1H, d, J = 7.6 Hz, -O<u>CH$_2$</u>OCH$_3$), 4.10 (1H, m, H17), 3.86 (1H, m, H'17), 3.73 (3H, s, -CO$_2$<u>CH$_3$</u>), 3.63 (1H, m, H3α), 3.38 (3H, s, -OCH$_2$O<u>CH$_3$</u>), 2.82 (1H, d, $J_{6,5}$ = 10.3 Hz, H6), 2.71 (1H, d overlapped, $J_{5,6}$ = 10.3 Hz, H5), 2.70 (1H, m overlapped, H11α), 2.40 (1H, m, H16), 2.18 (1H, m, H13), 1.76 (1H, d, $J_{14\alpha,14\beta}$ = 12.5 Hz, H14α), 1.13 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 177.9 (CO), 173.7 (CO), 96.2 (-O**C**H$_2$OCH$_3$), 94.9 (C10), 76.0 (C3), 74.6 (C12), 69.6 (C17), 56.2 (-OCH$_2$O**C**H$_3$), 53.5 (C4), 52.4 (-CO$_2$**C**H$_3$), 50.5 (C5), 45.9 (C6), 42.9 (C16), 41.9 (C13), 38.3 (C8), 31.1 (C9 or C14), 28.5 (C15), 27.7 (C9 or C14), 26.1 (C11), 25.1 (C2), 24.9 (C1), 14.6 (C18);

LRMS 404 ($M^{\cdot+}$, 17), 376 (25), 359 (8), 342 (10), 327 (5), 314 (12), 298 (46), 283 (9), 271 (9), 255 (11), 239 (27), 225 (100), 181 (32), 149 (72), 129 (29), 91 (80);

HRMS found 404.1835 ($M^{\cdot+}$), $C_{22}H_{28}O_7$ requires 404.1835.

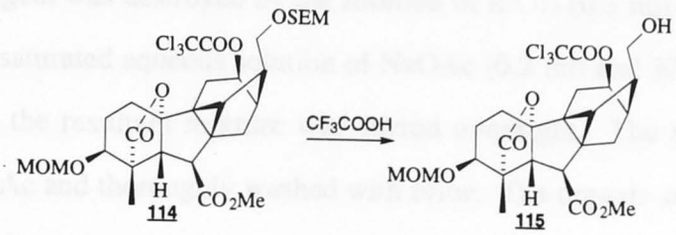
***ent*-10β-Hydroxy-12α-trichloroacetoxy-17-(2-trimethylsilylethoxymethoxy)-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>114</u>)**



Ether **<u>110</u>** (23 mg, 0.042 mmol) was dissolved in dry CH_2Cl_2 (2 ml). Et_3N (17 µl, 0.122 mmol) and CCl_3COCl (14 µl, 0.125 mmol) were added and the solution was allowed to stand at room temperature for 16 hours. The reaction mixture was diluted with EtOAc, washed with brine and dried. The solution was then concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 3:7) to afford the trichloroacetoxy derivative **<u>114</u>** (23 mg, 79%) as an oil:

^{1}H NMR (300 MHz, $CDCl_3$) 4.98 (1H, m, H12α), 4.71 (1H, d overlapped, J = 7.0 Hz, -O<u>C</u>H$_2$OCH$_3$), 4.70 (2H, s overlapped, -O<u>C</u>H$_2$OCH$_2$CH$_2$TMS), 4.60 (1H, d, J = 7.0 Hz, -O<u>C</u>H$_2$OCH$_3$), 3.79 (2H, d, J = 7.0 Hz, H17), 3.72 (3H, s, -CO$_2$<u>C</u>H$_3$), 3.60 (3H, m, -OCH$_2$O<u>C</u>H$_2$CH$_2$TMS, H3α), 3.38 (3H, s, -OCH$_2$O<u>C</u>H$_3$), 2.86 (1H, dd overlapped, $J_{11\alpha,11\beta}$ = 14.8 Hz, $J_{11\alpha,12\alpha}$ = 10.6 Hz, H11α), 2.85 (1H, d overlapped, $J_{6,5}$ = 9.2 Hz, H6), 2.79 (1H, d, $J_{5,6}$ = 9.2 Hz, H5), 2.42 (1H, m, H16), 2.28 (1H, m, H13), 2.07 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 12.4 Hz, $J_{14\beta,13}$ = 6.1 Hz, H14β), 1.75 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.4 Hz, H14α), 1.14 (3H, s, H18), 0.91 (2H, m, -OCH$_2$OCH$_2$<u>C</u>H$_2$TMS), 0.02 (3H, s, -OCH$_2$OCH$_2$CH$_2$<u>TMS</u>).

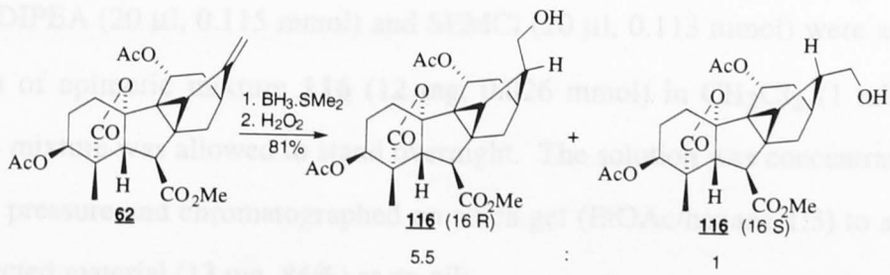
***ent*-17,10β-Dihydroxy-12α-trichloroacetoxy-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (115)**



SEM-ether **114** (9 mg, 0.013 mmol) was dissolved in CH_2Cl_2 (0.9 ml) and the solution was cooled to 0°C. CF_3COOH (1.8 ml) was added to the stirred solution dropwise over a period of 5 minutes and the mixture was allowed to stand at 0°C for 40 minutes. The liquid components were then removed (on a rotary evaporator) and the residue was dried under high vacuum. ^1H NMR analysis revealed the presence of the desired alcohol **115** (9 mg crude) contaminated by a small amount of the starting material:

^1H NMR (300 MHz, $CDCl_3$) 5.01 (1H, m, H12α), 4.71 (1H, d, J = 6.9 Hz, -O$\underline{CH_2}$OCH$_3$), 4.61 (1H, d, J = 6.9 Hz, -O$\underline{CH_2}$OCH$_3$), 3.91 (2H, m, H17), 3.73 (3H, s, -CO$_2\underline{CH_3}$), 3.64 (1H, m, H3α), 3.39 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.91 (1H, dd overlapped, $J_{11\alpha,11\beta}$ = 14.7 Hz, $J_{11\alpha,12\alpha}$ = 10.4 Hz, H11α), 2.86 (1H, d overlapped, $J_{6,5}$ = 9.2 Hz, H6), 2.79 (1H, d, $J_{5,6}$ = 9.2 Hz, H5), 2.42 (1H, m, H16), 2.35 (1H, m, H13), 2.06 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 12.6 Hz, $J_{14\beta,13}$ = 6.5 Hz, H14β), 1.78 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.6 Hz, H14α), 1.17 (3H, s, H18).

Hydroboration of olefin 62



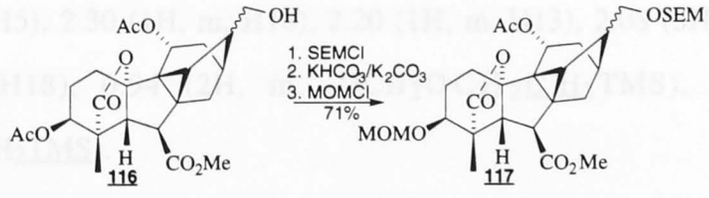
Compound **62** (20 mg, 0.045 mmol) was dissolved in dry THF (2 ml). A solution of $BH_3.SMe_2$ (0.5 ml, 25 µl of 10.2 M $BH_3.SMe_2$ solution diluted with 2 ml of

THF, approximately 0.065 mmol) was then added dropwise at room temperature. TLC analysis showed almost complete consumption of the starting material in 1 hour. The excess of the reagent was destroyed by the addition of EtOH (0.3 ml) and, after stirring for 5 minutes, a saturated aqueous solution of NaOAc (0.2 ml) and 30% H_2O_2 (0.3 ml) were added and the resultant mixture was stirred overnight. The mixture was then diluted with EtOAc and thoroughly washed with brine. The organic solution was dried, concentrated under reduced pressure and chromatographed on silica gel (EtOAc) to afford ***ent*-3α,11β-Diacetoxy-10β,17-dihydroxy-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** and its 16-epimer (**116**, 16*R*:16*S* = 5.5:1) as an inseparable mixture (oil, 17 mg, 81%):

major epimer: **^{1}H NMR** (300 MHz, $CDCl_3$) 5.79 (1H, dd, $J_{11\beta,12\beta}$ = 10.0 Hz, $J_{11\beta,12\alpha}$ = 3.6 Hz, H11β), 4.90 (1H, m, H3α), 3.73 (3H, s, -$CO_2\underline{CH_3}$), 3.69 (2H, m, H17), 2.93 (1H, d, $J_{6,5}$ = 9.4 Hz, H6), 2.63 (1H, d, $J_{5,6}$ = 9.4 Hz, H5), 2.26 (2H, m, H13, H16), 2.10 (6H, s, -OCO$\underline{CH_3}$), 1.03 (3H, s, H18);

minor epimer: **^{1}H NMR** (300 MHz, $CDCl_3$) 3.50 (d, J = 6.4 Hz, H17), 2.59 (d, $J_{5,6}$ = 9.3 Hz, H5), 1.06 (s, H18).

Protection of mixture 116 with SEMCl and replacement of the 3-acetate group to afford the MOM ether (117)



DIPEA (20 µl, 0.115 mmol) and SEMCl (20 µl, 0.113 mmol) were added to a solution of epimeric mixture **116** (12 mg, 0.026 mmol) in CH_2Cl_2 (1 ml) and the reaction mixture was allowed to stand overnight. The solution was concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 1:3) to afford the 17-protected material (13 mg, 84%) as an oil:

^{1}H NMR (major epimer, 300 MHz, $CDCl_3$) 5.80 (1H, dd, $J_{11\beta,12\beta}$ = 10.0 Hz, $J_{11\beta,12\alpha}$ = 3.4 Hz, H11β), 4.90 (1H, m, H3α), 4.68 (2H, s, -O$\underline{CH_2}$OCH_2CH_2TMS),

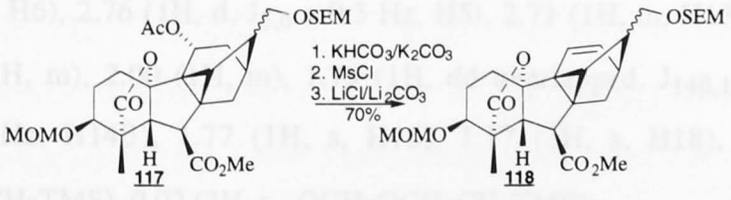
3.72 (3H, s, -$CO_2\underline{CH_3}$), 3.60 (2H, m, -$OCH_2O\underline{CH_2}CH_2TMS$), 3.52 (2H, m, H17), 2.93 (1H, d, $J_{6,5}$ = 9.3 Hz, H6), 2.62 (1H, d, $J_{5,6}$ = 9.3 Hz, H5), 2.29 (2H, m, H13, H16), 2.10 (6H, s, -$OCO\underline{CH_3}$), 1.01 (3H, s, H18), 0.94 (2H, m, -$OCH_2OCH_2\underline{CH_2}TMS$), 0.02 (3H, s, -$OCH_2OCH_2CH_2\underline{TMS}$).

This material (13 mg, 0.022 mmol) was dissolved in MeOH (1 ml) and the solution was treated with aqueous $KHCO_3/K_2CO_3$ (0.04 ml, 0.5 M, 50 mg $KHCO_3$/ 70 mg K_2CO_3 in 1 ml of solution). After being stirred at room temperature for 1 hour, the reaction mixture was poured into brine and the resulting mixture washed with EtOAc. The organic phase was dried and the solvent evaporated *in vacuo*.

The crude residue was redissolved in CH_2Cl_2 (1 ml). DIPEA (0.3 ml, excess) and MOMCl (0.1 ml, excess) were added followed by a catalytic amount of DMAP. TLC monitoring revealed that the reaction was complete after 24 hours. The solution was concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 2:3) to afford **117** (11 mg, 85%) as an oil:

major epimer: **^{1}H NMR** (300 MHz, $CDCl_3$) 5.79 (1H, dd, $J_{11\beta,12\beta}$ = 10.4 Hz, $J_{11\beta,12\alpha}$ = 3.4 Hz, H11β), 4.69 (1H, d overlapped, J = 6.9 Hz, -$O\underline{CH_2}OCH_3$), 4.68 (2H, s overlapped, -$O\underline{CH_2}OCH_2CH_2TMS$), 4.58 (1H, d, J = 6.9 Hz, -$O\underline{CH_2}OCH_3$), 3.72 (3H, s, -$CO_2\underline{CH_3}$), 3.59 (4H, m, -$OCH_2O\underline{CH_2}CH_2TMS$, H3α, H17), 3.48 (1H, m, H'17), 3.37 (3H, s, -$OCH_2O\underline{CH_3}$), 2.92 (1H, d, $J_{6,5}$ = 9.4 Hz, H6), 2.66 (1H, d, $J_{5,6}$ = 9.4 Hz, H5), 2.30 (1H, m, H16), 2.20 (1H, m, H13), 2.08 (3H, s, -$OCO\underline{CH_3}$), 1.11 (3H, s, H18), 0.94 (2H, m, -$OCH_2OCH_2\underline{CH_2}TMS$), 0.02 (3H, s, -$OCH_2OCH_2CH_2\underline{TMS}$).

Elimination of the 11-hydroxy group



Mixture **117** (11 mg, 0.019 mmol) was dissolved in MeOH (1 ml) and the solution was treated with aqueous $KHCO_3/K_2CO_3$ (0.2 ml, 0.5 M, 50 mg $KHCO_3$/

70 mg K_2CO_3 in 1 ml of solution). After stirring at room temperature for 12 hours, K_2CO_3 (10 mg, 0.072 mmol) was added to the solution and stirring was continued until TLC analysis showed that the starting material had almost disappeared (*ca* 30 hours). The solution was poured into brine, the resulting mixture extracted with EtOAc, the organic phase dried and the solvent removed.

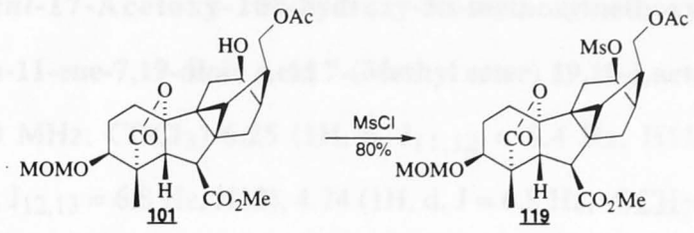
The crude product was redissolved in CH_2Cl_2 (1 ml). Et_3N (28 µl, 0.201 mmol) and MsCl (16 µl, 0.207 mmol) were added and the reaction mixture was allowed to stand overnight. The solution was diluted with EtOAc, subjected to standard work-up, dried and the solvent removed (on a rotary evaporator, then under high vacuum at 50°C).

The residue was redissolved in dry DMF (1 ml). Li_2CO_3 (30 mg) and LiCl (30 mg) were added and the mixture was stirred at 70°C overnight. The solvent was evaporated under high vacuum and the residue redissolved in EtOAc/brine. Both phases were thoroughly shaken in a separating funnel. The organic phase was dried, concentrated under reduced pressure and chromatographed on silica gel to afford ***ent*-10β-Hydroxy-3α–methoxymethoxy-17-(2-trimethylsilylethoxymethoxy)-20-nor-9α,15α-cyclogibberella-11-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** and its 16-epimer (**118**, 16*R*:16*S* = 5.5:1) as an inseparable mixture (oil, 7 mg, 70% from **117**):

major epimer: **^{1}H NMR** (300 MHz, $CDCl_3$) 6.22 (1H, dd, $J_{11,12}$ = 8.4 Hz, J = 1.0 Hz, H11), 5.73 (1H, dd, $J_{12,11}$ = 8.4 Hz, $J_{12,13}$ = 6.8 Hz, H12), 4.72 (1H, d, J = 7.0 Hz, -OCH_2OCH_3), 4.61 (1H, d, J = 7.0 Hz, -OCH_2OCH_3), 4.58 (2H, s, -OCH_2OCH_2CH_2TMS), 3.72 (3H, s, -CO_2CH_3), 3.67 (1H, m, H3α), 3.55 (2H, m, -OCH_2OCH_2CH_2TMS), 3.39 (3H, s, -OCH_2OCH_3), 3.14 (2H, m, H17), 2.91 (1H, d, $J_{6,5}$ = 9.3 Hz, H6), 2.76 (1H, d, $J_{5,6}$ = 9.3 Hz, H5), 2.71 (1H, m, H13), 2.23 (1H, m, H16), 2.12 (1H, m), 2.00 (1H, m), 1.79 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 11.7 Hz, $J_{14\beta,13}$ = 4.7 Hz, H14β), 1.77 (1H, s, H15), 1.17 (3H, s, H18), 0.91 (2H, m, -OCH_2OCH_2CH_2TMS), 0.02 (3H, s, -OCH_2OCH_2CH_2TMS);

minor epimer: **^{1}H NMR** (300 MHz, $CDCl_3$) 6.08 (m, H11, H12), 1.18 (s, H18).

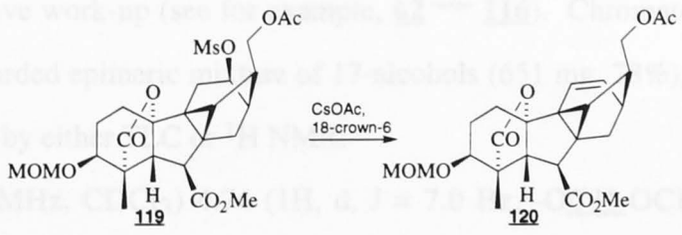
***ent*-17-Acetoxy-10β-hydroxy-3α-methoxymethoxy-12α-mesyloxy-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (119)**



Monoacetate **101** (6.4 mg, 0.014 mmol) was dissolved in CH_2Cl_2 (0.5 ml) and the solution was treated with Et_3N (20 µl, 0.143 mmol) and MsCl (10 µl, 0.129 mmol). After 16 hours at room temperature, chromatography of the solution on silica gel (EtOAc/hexane 1:1) afforded the 12-mesyloxy derivative **119** (6 mg, 80%) as an oil:

^{1}H NMR (300 MHz, $CDCl_3$) 4.81 (1H, m, H12α), 4.72 (1H, d, J = 6.7 Hz, $-O\underline{CH_2}OCH_3$), 4.61 (1H, d, J = 6.7 Hz, $-O\underline{CH_2}OCH_3$), 4.33 (2H, d, $J_{17,16}$ = 7.2 Hz, H17), 3.73 (3H, s, $-CO_2\underline{CH_3}$), 3.65 (1H, m, H3α), 3.39 (3H, s, $-OCH_2O\underline{CH_3}$), 2.98 (3H, s, $-OSO_2\underline{CH_3}$), 2.83 (1H, d overlapped, $J_{6,5}$ = 9.4 Hz, H6), 2.81 (1H, dd overlapped, H11α), 2.75 (1H, d, $J_{5,6}$ = 9.4 Hz, H5), 2.49 (1H, m, H16), 2.40 (1H, m, H13), 2.18 (1H, dd, $J_{11\beta,11\alpha}$ = 15.5 Hz, $J_{11\beta,112\alpha}$ = 5.5 Hz, H11β), 2.09 (3H, s, $OCO\underline{CH_3}$), 1.71 (1H, d, $J_{14\alpha,14\beta}$ = 11.6 Hz, H14α), 1.14 (3H, s, H18).

Reaction of mesylate 119 with CsOAc

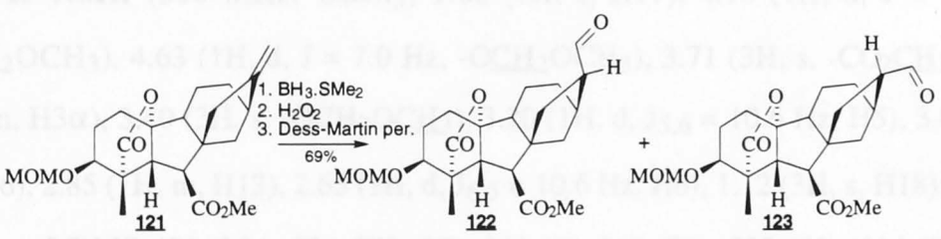


Mesylate **119** (5 mg, 0.009 mmol) was dissolved in dry toluene (1 ml) and the following components were added to the solution: dry CsOAc (17 mg, 0.089 mmol), dry 18-crown-6 ether (10 mg, 0.038 mmol) and dry powdered 4 Å molecular sieves (100 mg). The reaction mixture was heated at 60°C for 2 hours. TLC analysis indicated that only the starting material was present. The temperature was gradually elevated to 90°C and then to reflux. After 72 hours at reflux, the reaction mixture was

cooled, diluted with EtOAc, washed with brine, the organic phase dried and the solvent removed. NMR analysis of the crude product revealed the presence of the starting material and ***ent*-17-Acetoxy-10β-hydroxy-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberella-11-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone**:

^{1}H NMR (300 MHz, $CDCl_3$) 6.25 (1H, d, $J_{11,12}$ = 8.4 Hz, H11), 5.76 (1H, dd, $J_{12,11}$ = 8.4 Hz, $J_{12,13}$ = 6.8 Hz, H12), 4.74 (1H, d, J = 6.8 Hz, -O$\underline{CH_2}$OCH$_3$), 4.63 (1H, d, J = 6.8 Hz, -O$\underline{CH_2}$OCH$_3$), 3.72 (3H, s, -CO$_2\underline{CH_3}$), 3.39 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.92 (1H, d, $J_{6,5}$ = 9.6 Hz, H6), 2.77 (1H, d, $J_{5,6}$ = 9.6 Hz, H5), 2.05 (3H, s, OCO$\underline{CH_3}$), 1.15 (3H, s, H18), 0.95 (1H, d, $J_{14\alpha,14\beta}$ = 11.4 Hz, H14α).

Hydroboration of olefin <u>121</u> and oxidation of the resultant mixture of alcohols



Compound **<u>121</u>**[98] (800 mg, 2.05 mmol) was dissolved in dry THF (80 ml). $BH_3.SMe_2$ (0.5 ml of 10.2 M $BH_3.SMe_2$, approximately 5.27 mmol) was then added dropwise at room temperature. When TLC analysis showed almost complete consumption of the starting material (*ca* 2 hours), the reaction mixture was subjected to the usual oxidative work-up (see for example, **<u>62</u>** → **<u>116</u>**). Chromatography on silica gel (EtOAc) afforded epimeric mixture of 17-alcohols (651 mg, 78%), which could not be distinguished by either TLC or ^{1}H NMR:

^{1}H NMR (300 MHz, $CDCl_3$) 4.74 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.63 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 3.71 (3H, s, -CO$_2\underline{CH_3}$), 3.62 (3H, m, H17, H3α), 3.39 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.15 (1H, d, $J_{5,6}$ = 10.8 Hz, H5), 2.65 (1H, d, $J_{6,5}$ = 10.8 Hz, H6), 2.32 (1H, m overlapped, H16), 2.27 (1H, m overlapped, H13), 1.12 (3H, s, H18), 1.04 (1H, ddd, $J_{14\beta,14\alpha}$ = 13.0 Hz, $J_{14\beta,13}$ = 4.7 Hz, J = 2.2 Hz, H14β);

LRMS 408 ($M^{\cdot+}$, 13), 390 ($M^{\cdot+}$-H_2O, 4), 377 (14), 363 (8), 346 (45), 331 (18), 318 (100), 302 (51), 290 (43), 259 (29), 243 (80), 105 (41);

HRMS found 408.2147 ($M^{\cdot+}$), $C_{22}H_{32}O_7$ requires 408.2148.

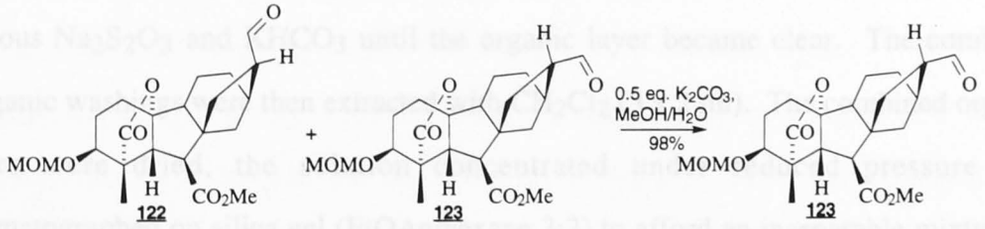
The mixture (60 mg, 0.147 mmol) was dissolved in CH_2Cl_2 (5 ml) and the solution was treated with the Dess-Martin periodinane (90 mg, 0.213 mmol). When TLC analysis indicated that the reaction was complete (*ca* 4 hours), the cloudy mixture was immediately poured into a separating funnel and washed successively with aqueous $Na_2S_2O_3$ and $KHCO_3$ until the organic layer became clear. The combined inorganic washings were then extracted with CH_2Cl_2 (3 x 3 ml). The combined organic layers were dried, the solution concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 2:3) to afford a mixture of ***ent*-10β-Hydroxy-3α-methoxymethoxy-17-oxo-20-norgibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone 122** and its 16–epimer **123** (**122** and **123** were obtained in the ratio of 2:1 and could be discriminated by TLC) as an oil (53 mg, 88%):

122: **^{1}H NMR** (300 MHz, $CDCl_3$) 9.80 (1H, s, H17), 4.74 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.63 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 3.71 (3H, s, -CO$_2\underline{CH_3}$), 3.64 (1H, m, H3α), 3.40 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.20 (1H, d, $J_{5,6}$ = 10.6 Hz, H5), 3.05 (1H, m, H16), 2.85 (1H, m, H13), 2.65 (1H, d, $J_{6,5}$ = 10.6 Hz, H6), 1.12 (3H, s, H18);

mixture: **LRMS** 406 ($M^{\cdot+}$, 22), 375 (20), 361 (8), 344 (31), 329 (23), 316 (97), 301 (59), 288 (100), 271 (23), 256 (30), 241 (73), 183 (62);

HRMS found 406.1991 ($M^{\cdot+}$), $C_{22}H_{30}O_7$ requires 406.1992.

Epimerisation of 122 with K_2CO_3



The mixture of **122** and **123** (52 mg, 0.128 mmol) was dissolved in MeOH (5 ml). H_2O (0.4 ml) and solid K_2CO_3 (8 mg, 0.056 mmol) were added and the solution was stirred at room temperature for 30 minutes at which stage TLC analysis indicated that only compound **123** was present in the reaction mixture. The solution was poured into brine and the resultant mixture was extracted with EtOAc. The organic phase was dried and the solvent removed. The residue contained pure **16-*epi*-**

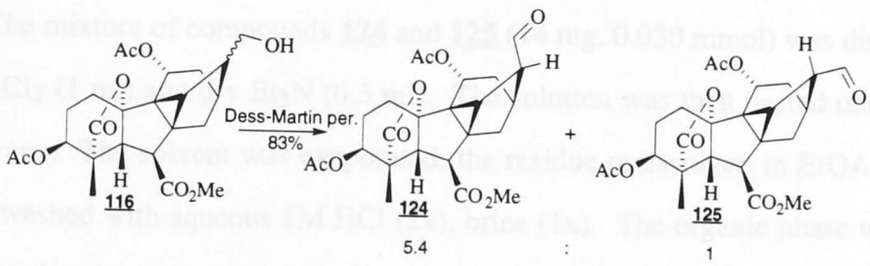
***ent*-10β-Hydroxy-3α-methoxymethoxy-17-oxo-20-norgibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**123**, 51 mg, 98%):

^{1}H NMR (300 MHz, $CDCl_3$) 9.65 (1H, d, J = 1.7 Hz, H17), 4.74 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.63 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 3.71 (3H, s, -CO$_2\underline{CH_3}$), 3.65 (1H, m, H3α), 3.40 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.20 (1H, d, $J_{5,6}$ = 10.6 Hz, H5), 2.70 (1H, d, $J_{6,5}$ = 10.6 Hz, H6), 2.47 (2H, m, H13, H16), 1.12 (3H, s, H18);

LRMS 406 (M·$^+$, 27), 378 (25), 361 (10), 344 (31), 329 (20), 316 (91), 301 (33), 288 (100), 271 (24), 256 (27), 241 (53), 183 (45);

HRMS found 406.1991 (M·$^+$), $C_{22}H_{30}O_7$ requires 406.1992.

Oxidation of mixture 116 with the Dess-Martin periodinane

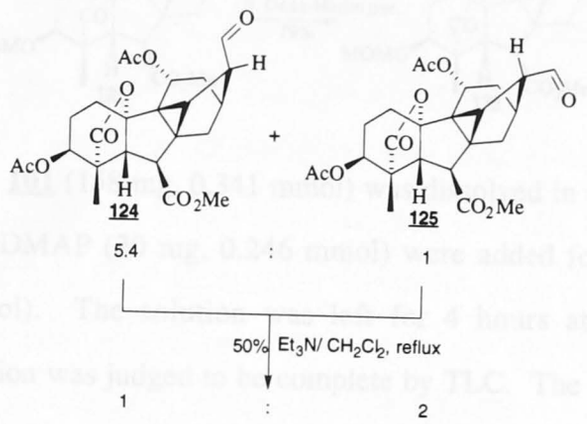


Epimeric mixture **116** (18 mg, 0.039 mmol) was dissolved in dry CH_2Cl_2 (2 ml) and the solution was treated with the Dess-Martin periodinane (38 mg, 0.090 mmol). When TLC analysis indicated that the reaction was complete (*ca* 5 hours), the cloudy mixture was immediately poured into a separating funnel and washed successively with aqueous $Na_2S_2O_3$ and $KHCO_3$ until the organic layer became clear. The combined inorganic washings were then extracted with CH_2Cl_2 (3 x 2 ml). The combined organic layers were dried, the solution concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 3:2) to afford an inseparable mixture of ***ent*-3α,11β-Diacetoxy-10β-hydroxy-17-oxo-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone 124** and its 16-epimer **125** (**124** : **125** = 5.4:1) as an oil (15 mg, 83%):

124: **^{1}H NMR** (300 MHz, $CDCl_3$) 9.88 (1H, s, H17), 5.70 (1H, dd, $J_{11\beta,12\beta}$ = 10.1 Hz, $J_{11\beta,12\alpha}$ = 3.5 Hz, H11β), 4.91 (1H, m, H3α), 3.73 (3H, s, -CO$_2\underline{CH_3}$), 2.98 (1H, m overlapped), 2.97 (1H, d overlapped, $J_{6,5}$ = 9.2 Hz, H6), 2.65 (1H, d, $J_{5,6}$ = 9.2 Hz, H5),

2.58 (1H, m), 2.35 (1H, m), 2.10 (3H, s, -OCO<u>CH</u>$_3$), 2.09 (3H, s, -OCO<u>CH</u>$_3$), 1.42 (1H, dm), 1.05 (3H, s, H18).

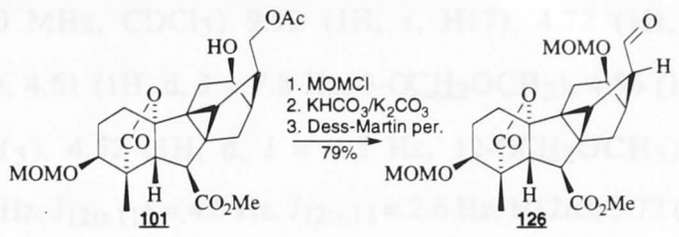
Et_3N-catalysed epimerisation of the mixture of <u>124</u> and <u>125</u>



The mixture of compounds **<u>124</u>** and **<u>125</u>** (14 mg, 0.030 mmol) was dissolved in dry CH_2Cl_2 (1 ml) and dry Et_3N (0.5 ml). The solution was then heated under reflux for 20 hours. The solvent was evaporated, the residue redissolved in EtOAc and the solution washed with aqueous 1M HCl (2x), brine (1x). The organic phase was dried, concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 3:2) to afford an inseparable mixture of ***ent*-3α,11β-Diacetoxy-10β-hydroxy-17-oxo-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone <u>124</u>** and its 16-epimer **<u>125</u>** (**<u>124</u>** : **<u>125</u>** = 1:2) as an oil (7 mg, 50%):

<u>125</u>: **^{1}H NMR** (300 MHz, $CDCl_3$) 9.65 (1H, d, J = 1.1 Hz, H17), 5.80 (1H, dd, $J_{11\beta,12\beta}$ = 10.0 Hz, $J_{11\beta,12\alpha}$ = 3.0 Hz, H11β), 4.91 (1H, m, H3α), 3.73 (3H, s, -CO_2<u>CH_3</u>), 2.96 (1H, d, $J_{6,5}$ = 9.3 Hz, H6), 2.78 (1H, d, $J_{5,6}$ = 9.3 Hz, H5), 2.54 (1H, m), 2.45 (1H, m), 2.26 (1H, m), 2.10 (3H, s, -OCO<u>CH_3</u>), 2.09 (3H, s, -OCO<u>CH_3</u>), 1.05 (3H, s, H18).

***ent*-10β-Hydroxy-3α,12α-bis-(methoxymethoxy)-17-oxo-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (126)**



Monoacetate **101** (158 mg, 0.341 mmol) was dissolved in dry CH_2Cl_2 (10 ml). DIPEA (1 ml) and DMAP (30 mg, 0.246 mmol) were added followed by MOMCl (1.2 ml, 15.80 mmol). The solution was left for 4 hours at room temperature whereupon the reaction was judged to be complete by TLC. The mixture was diluted with EtOAc, subjected to standard work-up, dried and the solvent removed under reduced pressure.

The crude product was redissolved in MeOH (15 ml) and the solution treated with aqueous $KHCO_3/K_2CO_3$ (1.0 ml, 0.5 M, 50 mg $KHCO_3$/70 mg K_2CO_3 in 1 ml of solution). The reaction mixture was then stirred at room temperature until TLC indicated that the hydrolysis was complete (*ca* 2 hours). The solution was poured into brine, the organic material extracted with EtOAc, the organic phase dried and the solvent evaporated *in vacuo.*

The crude alcohol was redissolved in dry CH_2Cl_2 (12 ml) and Dess-Martin periodinane (411 mg, 0.969 mmol) was added to the stirred solution. TLC analysis showed that the conversion of the 17-alcohol into the aldehyde **126** was complete in approximately 5 hours. The cloudy mixture was immediately poured into a separating funnel and washed successively with aqueous $Na_2S_2O_3$ and $KHCO_3$ until the organic layer became clear. The combined inorganic washings were then extracted with CH_2Cl_2 (3 x 5 ml). The combined organic layers were dried, concentrated under reduced pressure and the solution chromatographed on silica gel (EtOAc/hexane 1:1) to afford the aldehyde **126** (125 mg, 79%) as an oil, which crystallised from Et_2O:

mp 139-141°C;

$[\alpha]_D^{20}$ 86.5° (c 23.6 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2950, 1770, 1730, 1600, 1450, 1440, 1370, 1270, 1150, 1100, 1030 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 9.92 (1H, s, H17), 4.72 (1H, d, J = 7.8 Hz, 3-O$\underline{CH_2}OCH_3$), 4.61 (1H, d, J = 7.8 Hz, 3-O$\underline{CH_2}OCH_3$), 4.56 (1H, d, J = 7.5 Hz, 12-O$\underline{CH_2}OCH_3$), 4.52 (1H, d, J = 7.5 Hz, 12-O$\underline{CH_2}OCH_3$), 3.79 (1H, ddd, $J_{12\alpha,11\alpha}$ = 10.0 Hz, $J_{12\alpha,11\beta}$ = 4.8 Hz, $J_{12\alpha,13}$ = 2.6 Hz, H12α), 3.72 (3H, s, -$CO_2\underline{CH_3}$), 3.63 (1H, m, H3α), 3.38 (3H, s, -$OCH_2O\underline{CH_3}$), 3.29 (3H, s, -$OCH_2O\underline{CH_3}$), 2.88 (1H, m overlapped, H16), 2.85 (1H, d, $J_{6,5}$ = 9.1 Hz, H6), 2.77 (1H, d, $J_{5,6}$ = 9.1 Hz, H5), 2.64 (1H, m, H13), 2.56 (1H, dd, $J_{11\alpha,11\beta}$ = 14.4 Hz, $J_{11\alpha,12\alpha}$ = 10.0 Hz, H11α), 2.02 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 12.2 Hz, $J_{14\beta,13}$ = 6.3 Hz, H14β), 1.74 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.2 Hz, H14α), 1.15 (3H, s, H18);

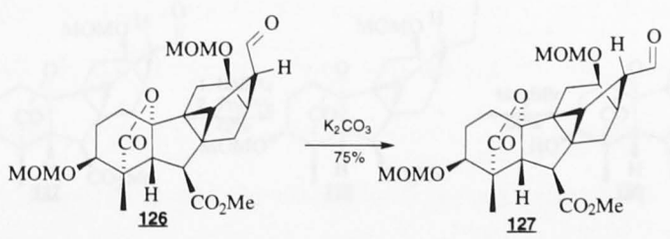
^{13}C NMR (75.5 MHz, $CDCl_3$) 201.7 (C17), 177.2 (CO), 172.8 (CO), 95.8 (-O$\mathbf{C}H_2CH_3$), 94.5 (-O$\mathbf{C}H_2CH_3$), 93.7 (C10), 75.7 (C3), 73.1 (C12), 55.7 (-$OCH_2\mathbf{O}CH_3$), 55.4 (-$OCH_2\mathbf{O}CH_3$), 53.2 (C4), 53.1 (C5), 52.0 (-$CO_2\mathbf{C}H_3$), 50.1 (C6), 45.0 (C16), 39.2 (C13), 38.0 (C8), 32.0 (C14 or C9), 31.5 (C14 or C9), 24.9 (CH_2), 24.6 (CH_2), 24.2 (CH_2), 21.6 (C15), 14.1 (C18);

LRMS 462 ($M^{\cdot+}$-2H, 3), 446 (3), 432 (7), 419 (13), 403 (18), 374 (92), 358 (38), 342 (12), 326 (22), 313 (100), 297 (54), 285 (33), 253 (34), 241 (44), 209 (86), 181 (80);

HRMS found 432.1783 ($M^{\cdot+}$-MeOH), $C_{23}H_{28}O_8$ requires 432.1784.

Anal. Found: C, 61.87; H, 6.87. Calcd for $C_{24}H_{32}O_9$: C, 62.06; H, 6.94.

16-*epi*-*ent*-10β-Hydroxy-3α,12α-bis-(methoxymethoxy)-17-oxo-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>127</u>)



K_2CO_3 (36 mg, 0.261 mmol) and H_2O (50 µl, 2.78 mmol) were added to a stirred solution of aldehyde **<u>126</u>** (123 mg, 0.265 mmol) in MeOH (10 ml). After being

stirred at room temperature for 4 hours, the solution was poured into brine, the resultant mixture extracted with EtOAc, the organic phase dried and concentrated under reduced pressure. Chromatography on silica gel (EtOAc/hexane 1:1) afforded the epimeric aldehyde **127** (92 mg, 75%) as an oil:

$[\alpha]_D^{20}$ 13.0° (c 21.3 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2940, 1770, 1720, 1450, 1435, 1360, 1260, 1150, 1100, 1040 cm^{-1};

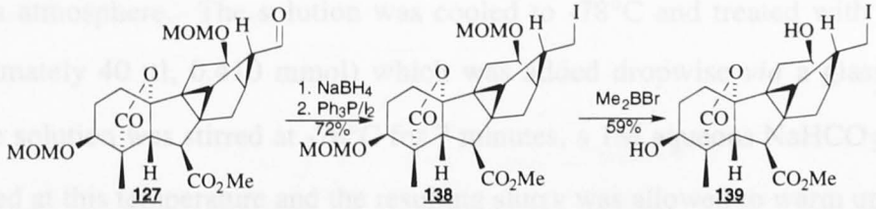
^{1}H NMR (300 MHz, $CDCl_3$) 9.67 (1H, s, H17), 4.72 (1H, d, J = 7.0 Hz, 3-O$\underline{CH_2}$OCH$_3$), 4.63 (2H, s overlapped, 12-O$\underline{CH_2}$OCH$_3$), 4.61 (1H, d overlapped, J = 7.0 Hz, 3-O$\underline{CH_2}$OCH$_3$), 3.80 (1H, m, H12α), 3.73 (3H, s, -CO$_2\underline{CH_3}$), 3.64 (1H, m, H3α), 3.38 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.35 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.04 (1H, s, H16), 2.87 (1H, d, $J_{6,5}$ = 9.1 Hz, H6), 2.82 (1H, d, $J_{5,6}$ = 9.1 Hz, H5), 2.58 (1H, dd overlapped, $J_{11\alpha,11\beta}$ = 14.4 Hz, $J_{11\alpha,12\alpha}$ = 9.6 Hz, H11α), 2.52 (1H, m overlapped, H13), 1.83 (1H, s overlapped, H15), 1.80 (1H, d, $J_{14\alpha,14\beta}$ = 12.6 Hz, H14α), 1.16 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 201.7 (C17), 177.2 (CO), 172.2 (CO), 95.8 (-O**C**H$_2$OCH$_3$), 95.0 (-O**C**H$_2$OCH$_3$), 93.5 (C10), 75.6 (C3), 73.0 (C12), 55.7 (-OCH$_2$O**C**H$_3$), 55.3 (-OCH$_2$O**C**H$_3$), 53.2 (C4), 52.0 (-CO$_2$**C**H$_3$), 49.7 (C5), 48.5 (C6), 44.8 (C16), 37.9 (C13), 37.7 (C8), 33.3 (C9), 28.4 (CH_2), 24.8 (CH_2), 24.4 (CH_2), 22.1 (C15), 14.1 (C18);

LRMS 464 ($M^{\cdot+}$, 7), 436 (10), 418 (3), 404 (18), 388 (22), 374 (100), 358 (30), 330 (38), 298 (49), 268 (46), 241 (35), 209 (35), 181 (48);

HRMS found 464.2048 ($M^{\cdot+}$), $C_{24}H_{32}O_9$ requires 464.2046.

Preparation of iodide 139



$NaBH_4$ (15 mg, 0.397 mmol) was added to a stirred solution of aldehyde **127** (91 mg, 0.196 mmol) in MeOH (5 ml). After the reaction mixture was stirred for

5 minutes, the excess of the reagent was destroyed with a few drops of aqueous 1M HCl. The solution was poured into brine, the resulting mixture extracted with EtOAc, the organic layer dried and the solvent removed under reduced pressure.

The crude alcohol was redissolved in dry CH_2Cl_2 (5 ml). PPh_3 (102 mg, 0.389 mmol) and imidazole (33 mg, 0.485 mmol) were added followed by a solution of iodine (99 mg, 0.390 mmol) in dry toluene (0.8 ml) which was added dropwise over a period of 5 minutes. The solution of iodine was immediately decolourized upon addition to the reaction mixture and a white precipitate was formed after 3 minutes. When TLC showed that the reaction was complete (*ca* 6 hours), the mixture was diluted with EtOAc, thoroughly washed with saturated aqueous $KHCO_3$ (1x), aqueous $Na_2S_2O_3$ (1x), H_2O, dried and concentrated under reduced pressure. Chromatography on silica gel (EtOAc/ hexane 2:3) afforded **16-*epi-ent*-10β-Hydroxy-3α,12α-bis(methoxymethoxy)-17-iodo-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone 138** (81 mg, 72%) as an oil:

^{1}H NMR (300 MHz, $CDCl_3$) 4.71 (1H, d, J = 7.3 Hz, 3-O$\underline{CH_2}$OCH$_3$), 4.62 (1H, d overlapped, J = 7.3 Hz, 3-O$\underline{CH_2}$OCH$_3$), 4.61 (1H, d overlapped, J = 6.7 Hz, 12-O$\underline{CH_2}$OCH$_3$), 4.58 (1H, d, J = 6.7 Hz, 12-O$\underline{CH_2}$OCH$_3$), 3.72 (1H, m overlapped, H12α), 3.72 (3H, s overlapped, -CO$_2\underline{CH_3}$), 3.63 (1H, m, H3α), 3.39 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.36 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.00 (2H, m, H17), 2.82 (1H, d, $J_{6,5}$ = 10.0 Hz, H6), 2.70 (1H, d, $J_{5,6}$ = 10.0 Hz, H5), 2.55 (1H, m overlapped, H13), 2.50 (1H, dd overlapped, $J_{11\alpha,11\beta}$ = 14.5 Hz, $J_{11\alpha,12\alpha}$ = 10.6 Hz, H11α), 1.80 (1H, dd, $J_{11\beta,11\alpha}$ = 14.5 Hz, $J_{11\beta,12\alpha}$ = 5.0 Hz, H11β), 1.43 (1H, d, $J_{14\alpha,14\beta}$ = 11.7 Hz, H14α), 1.18 (3H, s, H18).

Compound **138** (80 mg, 0.139 mmol) was dissolved in dry CH_2Cl_2 (5 ml) under an argon atmosphere. The solution was cooled to -78°C and treated with Me_2BBr (approximately 40 µl, 0.410 mmol) which was added dropwise *via* a glass pipette. After the solution was stirred at -78°C for 7 minutes, a 1M aqueous $NaHCO_3$ (0.5 ml) was added at this temperature and the resulting slurry was allowed to warm up to room temperature with efficient stirring. The mixture was then transferred into a separating funnel and washed with aqueous 1M $NaHCO_3$. The organic layer was separated and

the inorganic phase extracted with 3 x 2 ml of CH_2Cl_2. The combined organic extracts were dried, concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 3:2) to afford **16-*epi*-*ent*-3α,10β,12α-Trihydroxy-17-iodo-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone 129** (40 mg, 59%) as a solid, which crystallised from EtOAc/hexane:

mp 203-205°C;

$[\alpha]_D^{20}$ -24.9° (c 21.9 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 3600, 2950, 1770, 1730, 1450, 1430, 1260, 1170, 1140, 1000 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 3.92 (1H, m, H12α), 3.82 (1H, m, H3α), 3.72 (3H, s, $-CO_2\underline{CH_3}$), 3.06 (1H, dd, $J_{17,17}$ = 9.5 Hz, $J_{17,16}$ = 8.0 Hz, H17), 2.96 (1H, dd, $J_{17,17}$ = 9.5 Hz, $J_{17,16}$ = 8.0 Hz, H'17), 2.84 (1H, d, $J_{6,5}$ = 9.2 Hz, H6), 2.72 (1H, d, $J_{5,6}$ = 9.2 Hz, H5), 2.60 (1H, t, $J_{13,14\beta} = J_{13,16}$ = 7.7 Hz, H13), 2.51 (1H, dd, $J_{11\alpha,11\beta}$ = 14.4 Hz, $J_{11\alpha,12\alpha}$ = 9.5 Hz, H11α), 1.71 (1H, dd overlapped, $J_{11\beta,11\alpha}$ = 14.4 Hz, $J_{11\beta,12\alpha}$ = 4.5 Hz, H11β), 1.45 (1H, d, $J_{14\alpha,14\beta}$ = 12.2 Hz, H14α), 1.18 (3H, s, H18);

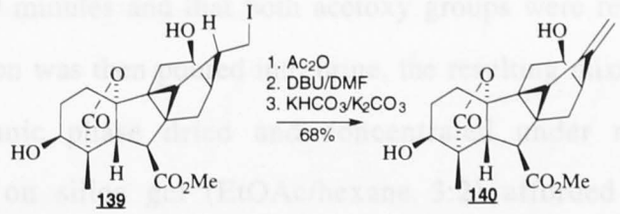
^{13}C NMR (75.5 MHz, $CDCl_3$) 177.6 (CO), 173.0 (CO), 94.1 (C10), 70.5 (C3), 68.3 (C12), 53.6 (C4), 52.2 ($-CO_2\mathbf{C}H_3$), 49.6 (C5), 45.5 (C6 or C16), 45.1 (C6 or C16), 39.1 (C13), 37.0 (C8), 34.7 (C9), 28.4 (C15), 27.9 (C14 or C11), 27.5 (C14 or C11), 26.4 (C2), 24.2 (C1), 14.3 (C18), 9.0 (C17);

LRMS 488 ($M^{\cdot+}$, 24), 470 ($M^{\cdot+}-H_2O$, 5), 426 (27), 410 (3), 385 (9), 361 (30), 343 (100), 299 (47), 281 (37), 255 (56), 239 (34), 221 (43), 195 (71);

HRMS found 488.0698 ($M^{\cdot+}$), $C_{20}H_{25}O_6I$ requires 488.0696.

Anal. Found: C, 49.07; H, 4.95. Calcd for $C_{20}H_{25}O_6I$: C, 49.19; H, 5.16.

Preparation of the 16-ene 140



Diol **139** (30 mg, 0.061 mmol) was dissolved in dry CH_2Cl_2 (2 ml). Acetic anhydride (35 µl, 0.371 mmol) and Et_3N (50 µl, 0.359 mmol) were added to the solution together with a small amount of DMAP (5 mg, 0.041 mmol). When TLC indicated that the reaction was complete, the solution was concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 35:65) to afford the protected material as an oil (33 mg, 94%):

^{1}H NMR (300 MHz, $CDCl_3$) 4.94 (1H, m, H3α), 4.77 (1H, m, H12α), 3.73 (3H, s, $-CO_2\underline{CH_3}$), 3.05 (1H, dd, $J_{17,17}$ = 9.7 Hz, $J_{17,16}$ = 7.7 Hz, H17), 2.92 (1H, dd, $J_{17,17}$ = 9.7 Hz, $J_{17,16}$ = 8.0 Hz, H'17), 2.85 (1H, d, $J_{6,5}$ = 8.9 Hz, H6), 2.65 (1H, d, $J_{5,6}$ = 8.9 Hz, H5), 2.55 (2H, m, H11α, H13), 2.21 (1H, m), 2.13 (3H, s, $-OCO\underline{CH_3}$), 2.04 (3H, s, $-OCO\underline{CH_3}$), 1.98 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 12.8 Hz, $J_{14\beta,13}$ = 6.5 Hz, H14β), 1.79 (1H, dd overlapped, $J_{11\beta,11\alpha}$ = 14.8 Hz, $J_{11\beta,12\alpha}$ = 4.4 Hz, H11β), 1.55 (1H, d, $J_{14\alpha,14\beta}$ = 12.8 Hz, H14α), 1.08 (3H, s, H18).

The diacetoxy derivative (33 mg, 0.058 mmol) was dissolved in dry DMF (2 ml) under an argon atmosphere. Powdered 4 Å molecular sieves were added to the solution and the heterogeneous mixture was stirred at room temperature for 1 hour. The temperature was then elevated to 50°C and DBU (90 µl, 0.581 mmol) was added dropwise to the mixture. The elimination process was monitored by TLC based on the different colour reactions of the starting material (blue) and the product (red). When TLC indicated that the reaction was complete (*ca* 2 days), the mixture was filtered through a short plug of silica gel using EtOAc as the eluent. The solvent was then removed under reduced pressure (on the rotary evaporator then under high vacuum) and the residue was redissolved in MeOH (2 ml). The solution was treated with aqueous $KHCO_3/K_2CO_3$ (0.12 ml, 0.5 M, 50 mg $KHCO_3$/70 mg K_2CO_3 in 1 ml of solution). TLC analysis revealed that one of the acetate groups (probably 3-OAc) was cleaved within the first 20 minutes and that both acetoxy groups were removed over a few hours. The solution was then poured into brine, the resulting mixture extracted with EtOAc, the organic phase dried and concentrated under reduced pressure. Chromatography on silica gel (EtOAc/hexane 3:2) afforded ***ent*-3α,10β,12α-Trihydroxy-20-nor-9α,15α-cyclogibberella-16-ene-7,19-dioic Acid 7-(Methyl ester)**

19,10-Lactone 140 (15 mg, 68% based on the diol **139**) as a solid, which crystallised from Et_2O/hexane:

mp 196-197°C;

$[\alpha]_D^{20}$ -24.7° (c 18.5 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 3610, 2940, 1760, 1730, 1660, 1450, 1370, 1290, 1140, 1050, 990 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 5.02 (1H, s, H17), 4.90 (1H, s, H'17), 3.88 (1H, m overlapped, H12α), 3.82 (1H, m overlapped, H3α), 3.72 (3H, s, $-CO_2\underline{CH_3}$), 2.88 (1H, d, $J_{6,5}$ = 9.2 Hz, H6), 2.79 (1H, d, $J_{5,6}$ = 9.2 Hz, H5), 2.64 (1H, dd, $J_{11\alpha,11\beta}$ = 14.8 Hz, $J_{11\alpha,12\alpha}$ = 9.1 Hz, H11α), 2.41 (1H, m, H13), 2.11 (1H, s, H15), 2.06 (1H, dd, $J_{14\beta,14\alpha}$ = 12.0 Hz, $J_{14\beta,13}$ = 6.4 Hz, H14β), 1.63 (1H, d, $J_{14\alpha,14\beta}$ = 12.0 Hz, H14α), 1.14 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 177.6 (CO), 172.6 (CO), 146.1 (C16), 107.0 (C17), 93.8 (C10), 70.3 (C3), 68.5 (C12), 53.5 (C4), 52.2 ($-CO_2\mathbf{C}H_3$), 49.2 (C5), 47.3 (C6), 45.4 (C13), 41.4 (C8), 37.1 (C9), 30.8 (C14), 30.3 (C15), 28.4 (C11 or C2), 27.9 (C11 or C2), 24.1 (C1), 14.0 (C18);

LRMS 360 ($M^{\cdot+}$, 21), 342 ($M^{\cdot+}-H_2O$, 14), 328 (36), 316 (9), 298 (24), 282 (14), 270 (39), 254 (78), 239 (37), 225 (14), 211 (18), 195 (100), 179 (28), 141 (35), 128 (30);

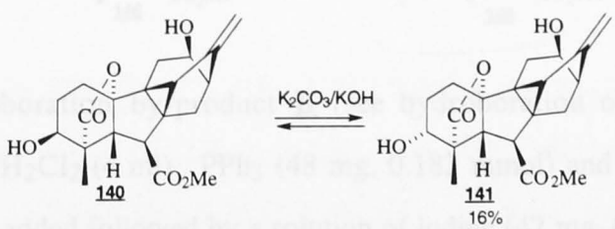
HRMS found 360.1574 ($M^{\cdot+}$), $C_{20}H_{24}O_6$ requires 360.1573.

Anal. Found: C, 65.61; H, 6.87. Calcd for $C_{20}H_{24}O_6$: C, 66.65; H, 6.71.

GC-MS (3,12-di-OTMS) 504 ($M^{\cdot+}$, 100), 488 (12), 472 (3), 445 (8), 429 (2), 414 (20), 401 (13), 370 (17), 355 (8), 311 (19), 285 (9), 254 (38), 221 (28), 195 (23), 129 (14);

KRI (3,12-di-OTMS) 2649.

Epimerisation of the 3-OH function



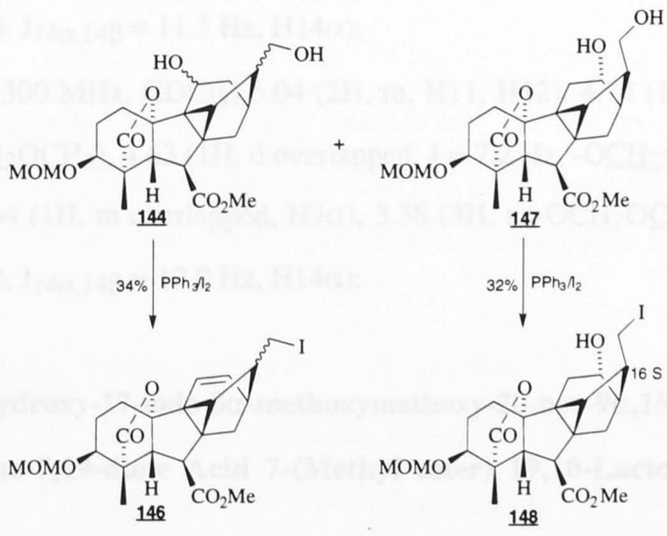
Diol **140** (11 mg, 0.031 mmol) was dissolved in MeOH (1.8 ml) and the solution was treated with aqueous K_2CO_3/KOH (0.36 ml, 1.38 g K_2CO_3 and 150 mg KOH in 10 ml of H_2O). After being stirred at room temperature for 24 hours, the solution was poured into brine and the resulting mixture extracted with EtOAc. The organic phase was dried, concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 3:2) to afford the recovered starting material (7 mg, 64%) followed by ***ent*-3β,10β,12α-Trihydroxy-20-nor-9α,15α-cyclogibberella-16-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone 141** (oil, 1.8 mg, 16%):

^{1}H NMR (300 MHz, $CDCl_3$) 5.01 (1H, s, H17), 4.91 (1H, s, H'17), 3.88 (1H, m, H12α), 3.74 (3H, s, $-CO_2\underline{CH_3}$), 3.60 (1H, m, H3β), 2.97 (1H, d, $J_{6,5}$ = 8.9 Hz, H6), 2.68 (1H, dd, $J_{11\alpha,11\beta}$ = 14.9 Hz, $J_{11\alpha,12\alpha}$ = 9.0 Hz, H11α), 2.41 (1H, m, H13), 2.10 (1H, d overlapped, $J_{5,6}$ = 8.9 Hz, H5), 2.00 (1H, s overlapped, H15), 1.19 (3H, s, H18);

GC-MS (3,12-di-OTMS) 504 ($M^{\cdot+}$, 81), 489 (8), 445 (9), 414 (18), 401 (15), 375 (12), 311 (12), 280 (14), 254 (88), 221 (100), 195 (61), 129 (38);

KRI (3,12-di-OTMS) 2777.

Reaction of mixture B with PPh_3/I_2



The hydroboration by-product **B** (see hydroboration of **96**, 60 mg) was dissolved in dry CH_2Cl_2 (4 ml). PPh_3 (48 mg, 0.183 mmol) and imidazole (15 mg, 0.220 mmol) were added followed by a solution of iodine (47 mg, 0.185 mmol) in dry

toluene (0.6 ml) which was added dropwise over a period of 5 minutes. The solution of iodine was immediately decolourized upon addition to the reaction mixture and a white precipitate was formed after 3 minutes. The mixture was then stirred at room temperature for 12 hours. Another portion of the reagents (imidazole, PPh_3 and I_2 in the same amounts) was added and the reaction mixture was stirred for a further 48 hours. The mixture was diluted with EtOAc, thoroughly washed with saturated aqueous $KHCO_3$ (1x), aqueous $Na_2S_2O_3$ (1x), H_2O, dried and the solution concentrated *in vacuo*. Chromatography on silica gel (EtOAc/hexane 1:3 then 1:1) gave in order of elution (given yields are based on the assumption that **B** was a mixture of diols **144** and **147** with the molecular weight of 422):

***ent*-10β-Hydroxy-17-iodo-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberella-11-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** and **16-*epi*-*ent*-10β-Hydroxy-17-iodo-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberella-11-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**146**, mixture of epimers, 25 mg, 34%):

16 *R*: **^{1}H NMR** (300 MHz, $CDCl_3$) 6.32 (1H, d, $J_{11,12}$ = 8.5 Hz, H11), 5.81 (1H, dd, $J_{12,11}$ = 8.5 Hz, $J_{12,13}$ = 6.8 Hz, H12), 4.74 (1H, d overlapped, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.62 (1H, d overlapped, J = 7.0 Hz, -OC$\underline{H_2}$OCH$_3$), 3.73 (3H, s, -CO$_2\underline{CH_3}$), 3.65 (1H, m overlapped, H3α), 3.39 (3H, s, -OCH$_2$O$\underline{CH_3}$), 1.15 (3H, s, H18), 1.02 (1H, d, $J_{14\alpha,14\beta}$ = 11.5 Hz, H14α);

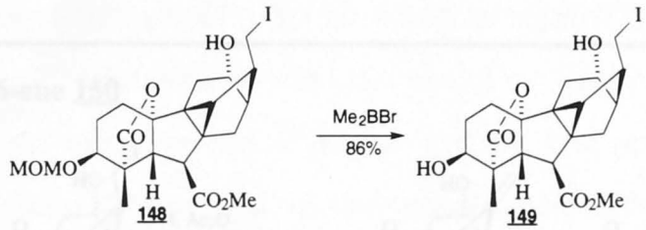
16 *S*: **^{1}H NMR** (300 MHz, $CDCl_3$) 6.04 (2H, m, H11, H12), 4.74 (1H, d overlapped, J = 7.2 Hz, -O$\underline{CH_2}$OCH$_3$), 4.63 (1H, d overlapped, J = 7.2 Hz, -O$\underline{CH_2}$OCH$_3$), 3.75 (3H, s, -CO$_2\underline{CH_3}$), 3.64 (1H, m overlapped, H3α), 3.38 (3H, s, -OCH$_2$O$\underline{CH_3}$), 1.20 (3H, s, H18), 0.83 (1H, d, $J_{14\alpha,14\beta}$ = 12.0 Hz, H14α);

***ent*-10β,12β-Dihydroxy-17-iodo-3α-methoxymethoxy-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** **148** (24 mg, 32%):

^{1}H NMR (300 MHz, $CDCl_3$) 4.71 (1H, d, J = 7.0 Hz, -OC$\underline{H_2}$OCH$_3$), 4.60 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.01 (1H, m, H12β), 3.72 (3H, s, -CO$_2\underline{CH_3}$), 3.63 (1H, m, H3α), 3.37 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.26 (1H, dd, $J_{17,17}$ = 9.8 Hz, $J_{17,16}$ = 7.5 Hz, H17),

3.06 (1H, t, $J_{17,17} = J_{17,16} = 9.8$ Hz, H'17), 2.81 (1H, d, $J_{6,5} = 9.2$ Hz, H6), 2.66 (1H, d, $J_{5,6} = 9.2$ Hz, H5), 2.56 (1H, m, H13), 2.42 (1H, dd, $J_{11\beta,11\alpha} = 15.3$ Hz, $J_{11\beta,12\beta} = 9.5$ Hz, H11β), 2.26 (1H, d, $J_{14\alpha,14\beta} = 12.3$ Hz, H14α), 2.16 (1H, m), 1.50 (1H, d, J = 3.3 Hz, H15), 1.14 (3H, s, H18).

***ent*-3α,10β,12β-Trihydroxy-17-iodo-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (149)**



Compound **148** (25 mg, 0.045 mmol) was dissolved in dry CH_2Cl_2 (2 ml) under an argon atmosphere. The solution was cooled to -78°C and treated with Me_2BBr (approximately 14 µl, 0.143 mmol) which was added dropwise *via* a glass pipette. After the solution was stirred at -78°C for 5 minutes, a 1M aqueous $NaHCO_3$ (0.3 ml) was added at this temperature and the resulting slurry was allowed to warm up to room temperature with efficient stirring. The mixture was then transferred into a separating funnel and washed with aqueous 1M $NaHCO_3$. The organic layer was separated and the inorganic phase extracted with 3 x 2 ml of CH_2Cl_2. The combined organic extracts were dried, concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 3:2) to afford the deprotected alcohol **149** (19 mg, 86%) as an oil:

IR ($CDCl_3$) ν_{max} 3610, 2950, 1770, 1730, 1450, 1440, 1370, 1260, 1170, 1050, 1010 cm^{-1};

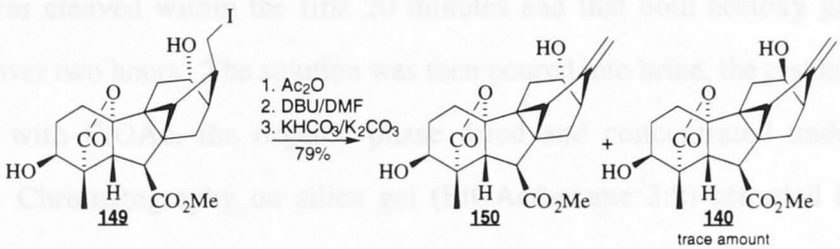
^{1}H NMR (300 MHz, $CDCl_3$) 4.01 (1H, m, H12β), 3.81 (1H, m, H3α), 3.72 (3H, s, $-CO_2\underline{CH_3}$), 3.25 (1H, dd, $J_{17,17'} = 9.8$ Hz, $J_{17,16} = 7.5$ Hz, H17), 3.06 (1H, t, $J_{17',17} = J_{17',16} = 9.8$ Hz, H'17), 2.83 (1H, d, $J_{6,5} = 9.2$ Hz, H6), 2.68 (1H, d, $J_{5,6} = 9.2$ Hz, H5), 2.55 (1H, m, H13), 2.43 (1H, dd, $J_{11\beta,11\alpha} = 15.2$ Hz, $J_{11\beta,12\beta} = 9.6$ Hz, H11β), 2.27 (1H, d, $J_{14\alpha,14\beta} = 12.2$ Hz, H14α), 1.49 (1H, d, J = 3.3 Hz, H15), 1.14 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 177.6 (CO), 173.1 (CO), 94.1 (C10), 70.4 (C3), 61.6 (C12), 53.4 (C4), 52.2 (-$CO_2\underline{C}H_3$), 48.8 (C5), 45.8 (C6), 44.4 (C16), 41.0 (C13), 38.9 (C8), 30.5 (C9), 27.9 (C14 or C11), 26.5 (C14 or C11), 25.5 (C2), 24.4 (C15), 24.2 (C1), 14.1 (C18), 5.4 (C17);

LRMS 488 ($M^{\cdot+}$, 100), 456 (5), 426 (81), 398 (7), 367 (9), 343 (52), 299 (68), 281 (92), 255 (53), 221 (74), 195 (78), 129 (54), 91 (31);

HRMS found 488.0698 ($M^{\cdot+}$), $C_{20}H_{25}O_6I$ requires 488.0696.

Preparation of 16-ene <u>150</u>



Diol **<u>149</u>** (19 mg, 0.039 mmol) was dissolved in dry CH_2Cl_2 (1.5 ml). Acetic anhydride (55 µl, 0.391 mmol) and Et_3N (54 µl, 0.387 mmol) were added to the solution together with a small amount of DMAP (2 mg, 0.016 mmol). When TLC indicated that the reaction was complete, the solution was concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 35:65) to afford the protected material as an oil (20 mg, 90%):

^{1}H NMR (300 MHz, $CDCl_3$) 4.95 (1H, m, H3α), 4.84 (1H, m, H12β), 3.74 (3H, s, -$CO_2\underline{CH_3}$), 3.26 (1H, m, H17), 3.12 (1H, m, H'17), 2.82 (1H, d, $J_{6,5}$ = 9.1 Hz, H6), 2.64 (1H, d, $J_{5,6}$ = 9.1 Hz, H5), 2.57 (1H, m overlapped, H13), 2.52 (1H, dd overlapped, $J_{11\beta,11\alpha}$ = 15.8 Hz, $J_{11\beta,12\beta}$ = 9.7 Hz, H11β), 2.27 (1H, m), 2.21 (1H, d, $J_{14\alpha,14\beta}$ = 12.4 Hz, H14α), 2.12 (3H, s, -OCO$\underline{CH_3}$), 2.05 (3H, s, -OCO$\underline{CH_3}$), 1.06 (3H, s, H18).

The diacetoxy derivative (20 mg, 0.035 mmol) was dissolved in dry DMF (1.5 ml) under an argon atmosphere. DBU (52 µl, 0.348 mmol) was added to the solution and the reaction mixture was stirred at 50°C. Similar to the synthesis of the 12-epimeric compound **<u>140</u>**, the elimination process was monitored by TLC based on

the different colour reactions of the starting material (blue) and the product (red). TLC after 2 hours indicated that the reaction had reached over 80% conversion. The reaction mixture was maintained at 50°C for another 5 hours in order to drive the elimination to completion. The solvent and the excess of the reagent were then removed under high vacuum and the residue was redissolved in EtOAc, the solution subjected to standard work-up, dried and the solvent evaporated *in vacuo*.

The crude product was redissolved in MeOH (1.5 ml) and the solution was treated with aqueous $KHCO_3/K_2CO_3$ (0.1 ml, 0.5 M, 50 mg $KHCO_3$/70 mg K_2CO_3 in 1 ml of solution). TLC analysis revealed that one of the acetate groups (probably 3-OAc) was cleaved within the first 20 minutes and that both acetoxy groups were removed over two hours. The solution was then poured into brine, the resulting mixture extracted with EtOAc, the organic phase dried and concentrated under reduced pressure. Chromatography on silica gel (EtOAc/hexane 3:2) afforded in order of elution:

***ent*-3α,10β,12β-Trihydroxy-20-nor-9α,15α-cyclogibberella-16-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**150**, 11 mg, 79% based on the diol) as a solid, which crystallised from EtOAc/hexane:

mp 241-242°C;

^{1}H NMR (300 MHz, $CDCl_3$) 4.86 (2H, s, H17), 3.93 (1H, m, H12β), 3.84 (1H, m, H3α), 3.73 (3H, s, $-CO_2\underline{CH_3}$), 2.94 (1H, d, $J_{6,5}$ = 9.2 Hz, H6), 2.74 (1H, d, $J_{5,6}$ = 9.2 Hz, H5), 2.52 (1H, t, $J_{13,14\beta} = J_{13,12\beta}$ = 4.7 Hz, H13), 2.42 (1H, dd, $J_{11\beta,11\alpha}$ = 14.8 Hz, $J_{11\beta,12\beta}$ = 9.4 Hz, H11β), 2.15 (1H, d, $J_{14\alpha,14\beta}$ = 11.9 Hz, H14α), 1.16 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 177.5 (CO), 172.7 (CO), 149.3 (C16), 105.3 (C17), 94.0 (C10), 70.5 (C3), 68.1 (C12), 53.4 (C4), 52.2 ($-CO_2\mathbf{C}H_3$), 48.5 (C5), 46.9 (C6), 45.9 (C13), 41.4 (C8), 37.0 (C9), 28.5 (C15), 28.0 (C14 or C11), 27.6 (C14 or C11), 25.4 (C2), 24.2 (C1), 14.1 (C18);

LRMS 360 ($M^{\cdot+}$, 27), 342 (18), 328 (38), 316 (10), 298 (31), 282 (16), 270 (33), 254 (72), 239 (34), 221 (29), 209 (16), 195 (100), 179 (21), 141 (31), 129 (28);

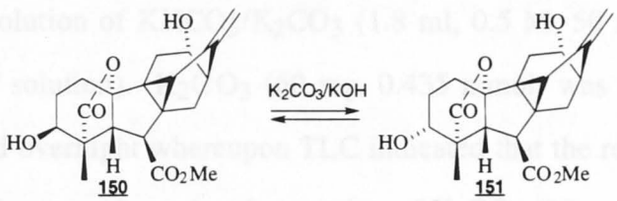
HRMS found 360.1574 ($M^{\cdot+}$), $C_{20}H_{24}O_6$ requires 360.1573.

GC-MS (3,12-di-OTMS) 504 ($M^{\cdot+}$, 100), 445 (8), 414 (18), 401 (13), 375 (15), 311 (8), 280 (13), 254 (81), 221 (95), 195 (57), 129 (35);

KRI (3,12-di-OTMS) 2614;

compound **140** (the precursor to this derivative was observed as a tiny amount of a contaminant in the ^{1}H NMR spectrum of iodide **148**) (0.5 mg), the ^{1}H NMR spectrum of which was identical with that of the previously prepared material.

Epimerisation of the 3-OH function



The same protocol as for the epimerisation of the 12β-alcohol **140** was followed; the starting material (10 mg, 0.028 mmol) was dissolved in MeOH (1.8 ml) and the solution was treated with 0.36 ml of aqueous K_2CO_3/KOH. Chromatography on silica gel (EtOAc/hexane 3:2) afforded the recovered starting material (6 mg, 60%) followed by ***ent*-3β,10β,12β-Trihydroxy-20-nor-9α,15α-cyclogibberella-16-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**151**, oil, 1.2 mg, 12%):

^{1}H NMR (300 MHz, $CDCl_3$) 4.84 (2H, s, H17), 3.91 (1H, m, H12β), 3.74 (3H, s, $-CO_2\underline{CH_3}$), 3.61 (1H, m, H3β), 3.01 (1H, d, $J_{6,5}$ = 8.7 Hz, H6), 2.52 (1H, t, $J_{13,14\beta} = J_{13,12\beta}$ = 4.8 Hz, H13), 2.39 (1H, dd, $J_{11\beta,11\alpha}$ = 14.8 Hz, $J_{11\beta,12\beta}$ = 9.4 Hz, H11β), 2.17 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.0 Hz, H14α), 2.06 (1H, d overlapped, $J_{5,6}$ = 8.7 Hz, H5), 1.86 (1H, s overlapped, H15), 1.19 (3H, s, H18);

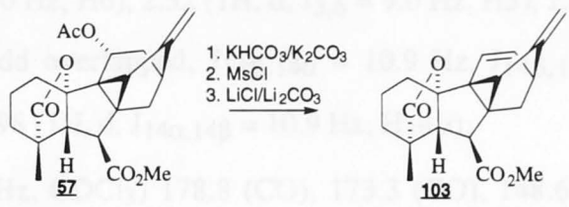
GC-MS (3,12-di-OTMS) 504 ($M^{\cdot+}$, 100), 489 (3), 457 (16), 445 (10), 414 (24), 401 (24), 375 (18), 285 (8), 254 (21), 221 (36), 195 (23), 129 (33);

KRI (3,12-di-OTMS) 2573.

5.3.3 3-deoxy series of 12-hydroxy-9,15-cyclo compounds

---

ent-10β-Hydroxy-20-nor-9α,15α-cyclogibberella-11,16-diene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (103)



Acetate **57** (230 mg, 0.596 mmol) was dissolved in MeOH (24 ml) and treated with an aqueous solution of $KHCO_3/K_2CO_3$ (1.8 ml, 0.5 M, 50 mg $KHCO_3$/70 mg K_2CO_3 in 1 ml of solution). K_2CO_3 (60 mg, 0.435 mmol) was then added and the mixture was stirred overnight whereupon TLC indicated that the reaction had reached approximately 70% conversion. Another portion of K_2CO_3 (80 mg, 0.580 mmol) was added and stirring was continued for a further 24 hours. The solution was poured into brine, the resulting mixture extracted with EtOAc, the organic layer dried and the solvent evaporated under reduced pressure.

The crude alcohol was redissolved in dry CH_2Cl_2 (10 ml) and Et_3N (0.6 ml, 4.3 mmol). After cooling in an ice bath, methanesulfonyl chloride (320 µl, 4.13 mmol) was added followed by DMAP (30 mg, 0.246 mmol) and the reaction mixture was allowed to warm to room temperature. TLC after 12 hours revealed that the reaction was complete. The solution was diluted with EtOAc, subjected to standard work-up, dried and the solvent evaporated *in vacuo*; the last traces of solvent were removed under high vacuum. The residue was redissolved in dry DMF (10 ml); dry LiCl (400 mg, 9.41 mmol) and dry Li_2CO_3 (400 mg, 5.41 mmol) were added and the reaction mixture was stirred at 80°C for 5 hours. The solvent was evaporated under high vacuum and the solid material was dissolved in EtOAc/brine. The organic phase was dried, concentrated under reduced pressure and chromatographed on silica gel to afford the desired diene **103** as a solid (135 mg, 70%), which crystallised from Et_2O:

mp 175-176°C;

$[\alpha]_D^{20}$ -210.4° (c 30.7 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2960, 1770, 1730, 1670, 1600, 1440, 1280, 1140 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 6.10 (1H, dd, $J_{11,12}$ = 8.3 Hz, J = 1.52 Hz, H11), 6.05 (1H, dd, $J_{12,11}$ = 8.3 Hz, $J_{12,13}$ = 6.5 Hz, H12), 4.84 (1H, s, H17), 4.69 (1H, s, H'17), 3.74 (3H, s, $-CO_2\underline{CH_3}$), 2.96 (1H, dd, $J_{13,12}$ = 6.5 Hz, $J_{13,14\beta}$ = 5.0 Hz, H13), 2.90 (1H, d, $J_{6,5}$ = 9.0 Hz, H6), 2.32 (1H, d, $J_{5,6}$ = 9.0 Hz, H5), 2.24 (1H, m), 2.08 (1H, s, H15), 1.87 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 10.9 Hz, $J_{14\beta,13}$ = 5.0 Hz, H14β), 1.12 (3H, s, H18), 0.98 (1H, d, $J_{14\alpha,14\beta}$ = 10.9 Hz, H14α);

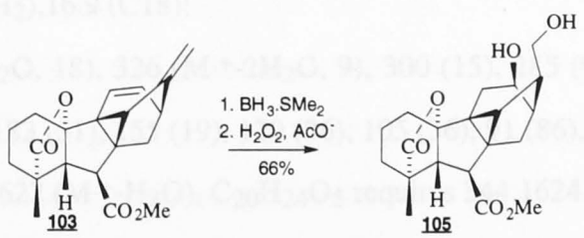
^{13}C NMR (75.5 MHz, $CDCl_3$) 178.8 (CO), 173.3 (CO), 148.6 (C16), 128.9 (C11), 121.5 (C12), 103.4 (C17), 92.2 (C10), 57.0 (C5), 52.5 ($-CO_2\mathbf{C}H_3$), 48.3 (C4), 45.5 (C6), 42.7 (C8), 41.9 (C13), 41.3 (C9), 35.3 (C14), 30.4 (C3 or C2), 29.1 (C3 or C2), 28.0 (C15), 19.5 (C1), 16.9 (C18);

LRMS 326 ($M^{\cdot+}$, 18), 282 (7), 267 (7), 223 (100), 239 (4), 207 (1), 193 (8), 181 (17), 167 (25), 152 (15), 128 (25), 115 (34);

HRMS found 326.1519 ($M^{\cdot+}$), $C_{20}H_{22}O_4$ requires 326.1518.

Anal. Found: C, 73.33; H, 6.82. Calcd for $C_{20}H_{22}O_4$: C, 73.60; H, 6.79.

***ent*-10β,12α,17-Trihydroxy-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>105</u>)**



Diene **<u>103</u>** (130 mg, 0.399 mmol) was dissolved in dry THF (26 ml) under Ar. The solution was cooled to 0°C and treated with $BH_3.SMe_2$ (3 ml, approximately 0.620 mmol; 160 µl of 10.2 M $BH_3.SMe_2$ diluted with 8 ml of dry THF) which was added dropwise over a period of 10 minutes. The reaction mixture was allowed to warm to room temperature over 3 hours at which stage TLC indicated that the starting material had almost disappeared. The excess of the reagent was then destroyed by the addition of EtOH (1 ml) and, after stirring for 5 minutes, the solution was subjected to oxidative work-up: saturated aqueous NaOAc (1 ml) was added followed by 30% H_2O_2

(1.5 ml) and the resulting mixture was stirred for 24 hours. K_2CO_3 (60 mg, 0.435 mmol) was added after this period and stirring was continued for another 24 hours. The solution was concentrated under reduced pressure, diluted with EtOAc and thoroughly washed with brine. The organic phase was dried, concentrated *in vacuo* and chromatographed on silica gel (EtOAc/MeOH 98:2) to afford diol **105** (95 mg, 66%) as an oil, which crystallised from EtOAc/Et_2O:

mp 196-197°C;

$[\alpha]_D^{20}$ -7.3° (c 28.9 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 3620, 3480, 2960, 1760, 1730, 1440, 1270, 1200, 1180, 1140, 1060 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 3.97 (1H, m, H12α), 3.85 (2H, m, H17), 3.72 (3H, s, -$CO_2\underline{CH_3}$), 2.83 (1H, d, $J_{6,5}$ = 8.8 Hz, H6), 2.65 (1H, dd, $J_{11\alpha,11\beta}$ = 14.4 Hz, $J_{11\alpha,12\alpha}$ = 10.3 Hz, H11α), 2.34 (1H, m, H16), 2.12 (1H, m overlapped, H13), 2.08 (1H, d overlapped, $J_{5,6}$ = 8.8 Hz, H5), 2.00 (1H, m), 1.61 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.2 Hz, H14α), 1.31 (1H, d, J = 3.3 Hz, H15), 1.08 (3H, s, H18);

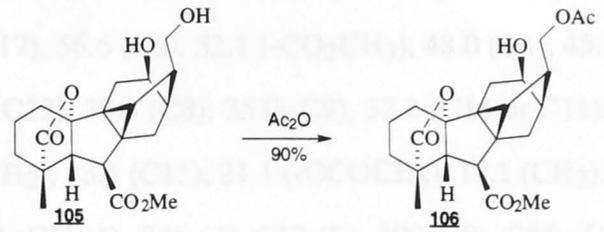
^{13}C NMR (75.5 MHz, $CDCl_3$) 179.3 (CO), 173.5 (CO), 94.0 (C10), 68.9 (C12), 62.8 (C17), 57.0 (C5), 52.4 (-$CO_2\mathbf{C}H_3$), 48.3 (C4), 45.6 (C6), 42.8 (C16), 38.8 (C13), 38.0 (C8), 35.2 (C9), 32.5 (C14 or C11), 32.3 (C14 or C11), 27.7 (CH_2), 27.3 (CH_2), 23.2 (C15), 19.3 (CH_2),16.9 (C18);

LRMS 344 ($M^{\cdot+}$-H_2O, 18), 326 ($M^{\cdot+}$-$2H_2O$, 9), 300 (15), 285 (9), 266 (27), 255 (7), 241 (30), 227 (23), 183 (51), 155 (19), 129 (36), 105 (36), 91 (86), 55 (100);

HRMS found 344.1622 ($M^{\cdot+}$-H_2O), $C_{20}H_{24}O_5$ requires 344.1624.

Anal. Found: C, 66.22; H, 7.55. Calcd for $C_{20}H_{26}O_6$: C, 66.28; H, 7.23.

***ent*-17-Acetoxy-10β,12α-dihydroxy-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (106)**



Diol **105** (90 mg, 0.249 mmol) was dissolved in dry CH_2Cl_2 (18 ml). Dry Et_3N (70 µl, 0.502 mmol) was added followed by Ac_2O (35 µl, 0.371 mmol) and the solution was left to stand at room temperature. TLC analysis after three days indicated that most of the starting material remained unreacted and another portion of Et_3N (70 µl, 0.502 mmol) and Ac_2O (35 µl, 0.371 mmol) was therefore added. More Ac_2O (10 µl) and Et_3N (35 µl) was added after 72 hours and the reaction was allowed to proceed for another 24 hours. At this stage TLC monitoring showed that the major product was the desired 17-monoacetate **106**, while the amounts of the starting material and the 12,17-diacetate were roughly equal. The solution was concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 1:1 then 7:3) to afford the 12,17-diacetate (22 mg) and the 17-monoacetate **106** (60 mg). Continuing elution with EtOAc/MeOH 98:2 gave the recovered diol **105** (20 mg). The 12,17-diacetoxy compound was, without characterization, hydrolysed into the diol **105** with aqueous $KHCO_3/K_2CO_3$ (see **100** → **101**) and together with the recovered starting material re-treated under the above conditions. After three cycles, the 17-monoacetate **106** (total product: 90 mg, 90%) was obtained as an oil, which crystallised from Et_2O at -30°C:

mp 169-170°C;

$[\alpha]_D^{20}$ 10.2° (c 26.0 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 3610, 3400, 2960, 1770, 1730, 1440, 1370, 1270, 1140 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 4.41 (2H, m, H17), 3.97 (1H, m, H12α),, 3.71 (3H, s, $-CO_2\underline{CH_3}$), 2.82 (1H, d, $J_{6,5}$ = 8.9 Hz, H6), 2.63 (1H, dd, $J_{11\alpha,11\beta}$ = 14.4 Hz, $J_{11\alpha,12\alpha}$ = 10.3 Hz, H11α), 2.35 (1H, m, H16), 2.09 (1H, d overlapped, $J_{5,6}$ = 8.9 Hz,

H5), 2.06 (3H, s overlapped, -OCOCH$_3$), 1.60 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.0 Hz, H14α), 1.07 (3H, s, H18);

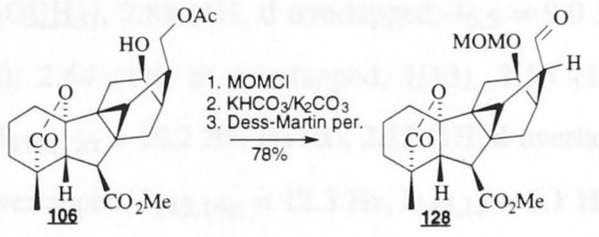
^{13}C NMR (75.5 MHz, $CDCl_3$) 178.7 (CO), 173.2 (CO), 171.1 (CO), 93.4 (C10), 69.2 (C12), 65.2 (C17), 56.6 (C5), 52.1 (-CO_2**C**H$_3$), 48.0 (C4), 45.3 (C6), 40.3 (C16 or C13), 39.8 (C16 or C13), 37.8 (C8), 35.0 (C9), 32.2 (C14 or C11), 32.1 (C14 or C11), 27.1 (CH_2), 26.8 (CH_2), 23.1 (C15), 21.1 (-OCO**C**H$_3$), 19.1 (CH_2), 16.7 (C18);

LRMS 344 (M·$^+$-AcOH, 4), 326 (3), 312 (8), 300 (19), 285 (5), 266 (7), 254 (60), 240 (62), 223 (17), 211 (10), 183 (32), 155 (35), 141 (54), 115 (51), 91 (100);

HRMS found 344.1622 (M·$^+$-AcOH), $C_{20}H_{24}O_5$ requires 344.1624.

Anal. Found: C, 65.50; H, 7.13. Calcd for $C_{22}H_{28}O_7$: C, 65.33; H, 6.98.

***ent*-10β-Hydroxy-12α-methoxymethoxy-17-oxo-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>128</u>)**



Monoacetate **<u>106</u>** (90 mg, 0.223 mmol) was dissolved in dry CH_2Cl_2 (5 ml). DIPEA (1 ml, 5.74 mmol) and DMAP (10 mg, 0.082 mmol) were added to the solution followed by MOMCl (0.5 ml, 6.58 mmol). The solution was left overnight, whereupon the reaction was judged to be complete by TLC. The mixture was diluted with EtOAc, subjected to standard work-up, dried and the solvent removed under reduced pressure.

The crude product was redissolved in MeOH (9 ml) and the solution treated with aqueous $KHCO_3/K_2CO_3$ (0.5 ml, 0.5 M, 50 mg $KHCO_3$/70 mg K_2CO_3 in 1 ml of solution). More reagent (0.2 ml of the above carbonate solution) was added after 2 hours and stirring was continued until TLC indicated that the hydrolysis was complete (*ca* 1 hour). The reaction mixture was poured into brine, the resulting mixture extracted with EtOAc, the organic phase dried and the solvent evaporated *in vacuo*.

The crude alcohol was redissolved in dry CH_2Cl_2 (7 ml) and Dess-Martin periodinane (400 mg, 0.943 mmol) was added to the stirred solution. TLC analysis

showed that the conversion of the 17-alcohol into aldehyde **128** was complete in 4 hours. The cloudy mixture was immediately poured into a separating funnel and washed successively with aqueous $Na_2S_2O_3$ and $KHCO_3$ until the organic layer became clear. The combined inorganic washings were then extracted with CH_2Cl_2 (3 x 5 ml). The combined organic extracts were dried, concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 1:1) to afford aldehyde **128** (70 mg, 78%) as a crystalline solid, which was recrystallised from Et_2O:

mp 113-114°C;

$[\alpha]_D^{20}$ 57.2° (c 25.1 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2960, 1770, 1730, 1720, 1440, 1380, 1270, 1200, 1150, 1040 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 9.91 (1H, d, J = 1.0 Hz, H17), 4.56 (1H, d, J = 6.8 Hz, -OC$\underline{H_2}$OCH$_3$), 4.52 (1H, d, J = 6.8 Hz, -OC$\underline{H_2}$OCH$_3$), 3.79 (1H, ddd, $J_{12\alpha,11\alpha}$ = 10.2 Hz, $J_{12\alpha,11\beta}$ = 4.8 Hz, $J_{12\alpha,13}$ = 2.6 Hz, H12α), 3.72 (3H, s, -$CO_2\underline{CH_3}$), 3.27 (3H, s, -$OCH_2O\underline{CH_3}$), 2.88 (1H, d overlapped, $J_{6,5}$ = 9.0 Hz, H6), 2.85 (1H, m overlapped, H16), 2.64 (1H, m overlapped, H13), 2.56 (1H, dd overlapped, $J_{11\alpha,11\beta}$ = 14.7 Hz, $J_{11\alpha,12\alpha}$ = 10.2 Hz, H11α), 2.12 (1H, d overlapped, $J_{5,6}$ = 9.0 Hz, H5), 2.02 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 12.3 Hz, $J_{14\beta,13}$ = 6.1 Hz, H14β), 1.81 (1H, dd overlapped, $J_{11\beta,11\alpha}$ = 14.7 Hz, $J_{11\beta,12\alpha}$ = 4.8 Hz, H11β), 1.73 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.3 Hz, H14α), 1.09 (3H, s, H18);

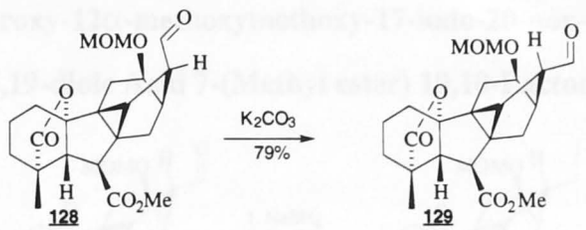
^{13}C NMR (75.5 MHz, $CDCl_3$) 201.7 (C17), 178.4 (CO), 172.9 (CO), 94.5 (-OCH_2OCH_3), 93.2 (C10), 73.0 (C12), 56.6 (C5), 55.4 (-$OCH_2O\mathbf{C}H_3$), 53.1 (C6), 52.0 (-$CO_2\mathbf{C}H_3$), 47.9 (C4), 45.3 (C16), 39.2 (C13), 37.9 (C8), 35.0 (C9), 32.3 (C14 or C11), 31.5 (C14 or C11), 26.9 (CH_2), 24.9 (CH_2), 21.6 (C15), 18.9 (CH_2), 16.6 (C18);

LRMS 404 ($M^{\cdot+}$, 1), 386 (0.6), 372 (1), 359 (23), 328 (16), 314 (70), 299 (14), 285 (10), 270 (17), 255 (19), 241 (43), 227 (43), 183 (58), 155 (40), 129 (37), 91 (68), 55 (100);

HRMS found 404.1835 ($M^{\cdot+}$), $C_{22}H_{28}O_7$ requires 404.1835.

Anal. Found: C, 64.90; H, 7.05. Calcd for $C_{22}H_{28}O_7$: C, 65.33; H, 6.98.

16-*epi*-*ent*-10β-Hydroxy-12α-methoxymethoxy-17-oxo-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (127)



K_2CO_3 (6 mg, 0.043 mmol) was added to a stirred solution of aldehyde **128** (68 mg, 0.168 mmol) in MeOH (7 ml) and H_2O (0.24 ml, 13.3 mmol). TLC analysis after 5 hours indicated that the reaction had reached approximately 50% conversion. More reagent (18 mg, 0.130 mmol) and H_2O (0.35 ml, 19.4 mmol) was added and stirring was continued for another 1.5 hours whereupon the epimerisation was judged to be complete by TLC. The reaction mixture was diluted with EtOAc, washed with brine, dried and concentrated *in vacuo*. Chromatography on silica gel (EtOAc/hexane 1:1) afforded the 17R aldehyde **129** (54 mg, 79%) as a crystalline solid, which was recrystallised from Et_2O:

mp 111-113°C;

$[\alpha]_D^{20}$ -34.5° (c 25.5 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2960, 1770, 1730, 1440, 1380, 1260, 1150, 1040 cm^{-1};

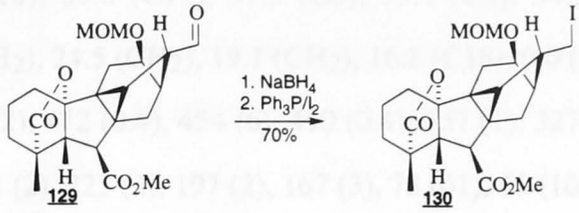
^{1}H NMR (300 MHz, $CDCl_3$) 9.65 (1H, s, H17), 4.62 (2H, s, -O$\underline{CH_2}$OCH$_3$), 3.79 (1H, m, H12α), 3.73 (3H, s, -CO$_2\underline{CH_3}$), 3.35 (3H, s, -OCH$_2$O$\underline{CH_3}$), 3.04 (1H, s, H16), 2.85 (1H, d, $J_{6,5}$ = 8.8 Hz, H6), 2.59 (1H, dd, $J_{11\alpha,11\beta}$ = 14.5 Hz, $J_{11\alpha,12\alpha}$ = 9.7 Hz, H11α), 2.51 (1H, m, H13), 2.18 (1H, d, $J_{5,6}$ = 8.8 Hz, H5), 2.02 (1H, m), 1.50 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.6 Hz, H14α), 1.10 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 201.6 (C17), 178.5 (CO), 172.3 (CO), 95.1 (-O**C**H$_2$OCH$_3$), 93.1 (C10), 73.0 (C12), 56.3 (C5), 55.4 (-OCH$_2$O**C**H$_3$), 52.1 (-CO$_2$**C**H$_3$), 48.5 (C6), 47.9 (C4), 45.1 (C16), 38.0 (C13), 37.6 (C8), 35.0 (C9), 33.7 (C14), 28.4 (C11), 27.1 (CH_2), 24.8 (CH_2), 22.0 (C15), 19.0 (CH_2), 16.6 (C18);

LRMS 342 (2), 328 (2), 314 (3), 298 (7), 283 (2), 270 (4), 255 (5), 241 (8), 227 (7), 183 (41), 155 (34), 141 (61), 128 (62), 115 (70), 91 (100);

Anal. Found: C, 65.15; H, 7.29. Calcd for $C_{22}H_{28}O_7$: C, 65.33; H, 6.98.

16-*epi-ent*-10β-Hydroxy-12α-methoxymethoxy-17-iodo-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (130)



$NaBH_4$ (15 mg, 0.397 mmol) was added to a solution of aldehyde **129** (54 mg, 0.134 mmol) and the reaction mixture was stirred at room temperature for 5 minutes. The excess of the reagent was destroyed by the addition of a few drops of aqueous 1M HCl and the solution was poured into brine. The resulting mixture was extracted with EtOAc, the organic layer dried and the solvent evaporated.

The crude alcohol was redissolved in dry CH_2Cl_2 (2.5 ml). PPh_3 (97 mg, 0.370 mmol) and imidazole (30 mg, 0.441 mmol) were added followed by a solution of iodine (75 mg, 0.295 mmol) in dry toluene (0.6 ml) which was added dropwise over a period of 5 minutes. The solution of iodine was immediately decolourized upon addition to the reaction mixture and a white precipitate was formed after 3 minutes. When TLC analysis showed that the reaction was complete (*ca* 3 hours), the mixture was diluted with EtOAc, thoroughly washed with saturated aqueous $KHCO_3$ (1x), aqueous $Na_2S_2O_3$ (1x), H_2O, dried and concentrated *in vacuo*. Chromatography on silica gel (EtOAc/hexane 3:7) afforded iodide **130** (48 mg, 70%) as an oil:

$[\alpha]_D^{20}$ -19.2° (c 19.8 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2960, 1770, 1730, 1440, 1260, 1180, 1150, 1040 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 4.64 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 4.58 (1H, d, J = 7.0 Hz, -O$\underline{CH_2}$OCH$_3$), 3.73 (3H, s overlapped, -CO$_2\underline{CH_3}$), 3.72 (1H, m overlapped, H12α), 3.35 (3H, s, -OCH$_2$O$\underline{CH_3}$), 2.98 (2H, m, H17), 2.86 (1H, d, $J_{6,5}$ = 8.8 Hz, H6), 2.54 (1H, m overlapped, H13), 2.50 (1H, dd overlapped, $J_{11\alpha,11\beta}$ = 14.4 Hz, $J_{11\alpha,12\alpha}$ = 9.8 Hz, H11α), 2.17 (1H, m, H16), 2.07 (1H, d overlapped, $J_{5,6}$ = 8.8 Hz, H5), 1.93 (1H, dd, $J_{14\beta,14\alpha}$ = 12.6 Hz, $J_{14\beta,13}$ = 6.4 Hz, H14β), 1.79 (1H,

dd overlapped, $J_{11\beta,11\alpha}$ = 14.4 Hz, $J_{11\beta,12\alpha}$ = 4.6 Hz, H11β), 1.43 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.6 Hz, H14α), 1.11 (3H, s, H18);

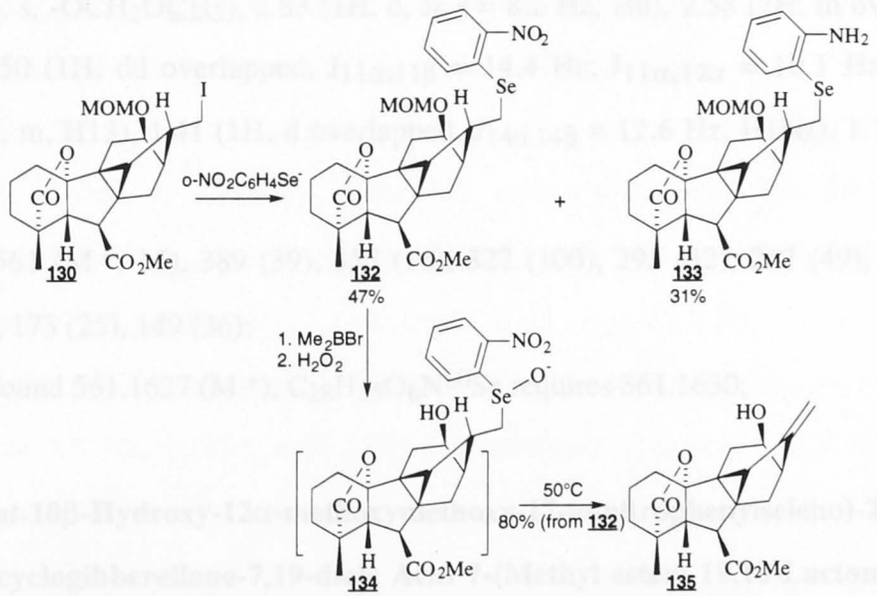
^{13}C NMR (75.5 MHz, $CDCl_3$) 178.5 (CO), 173.1 (CO), 94.7 (-OCH_2OCH$_3$), 93.3 (C10), 73.1 (C12), 56.8 (C5), 55.5 (-OCH$_2$OCH_3), 52.1 (-CO$_2$CH_3), 48.0 (C4), 45.4 (C6), 42.4 (C16), 39.5 (C13), 37.3 (C8), 35.1 (C9), 34.9 (C14), 28.3 (C15), 27.4 (C11), 27.2 (CH_2), 24.5 (CH_2), 19.1 (CH_2), 16.8 (C18), 9.0 (C17);

LRMS 516 ($M^{\cdot+}$, 0.2), 472 (0.4), 454 (6), 410 (0.4), 357 (1), 327 (2), 313 (1), 299 (1), 267 (2), 255 (1), 241 (2), 223 (2), 197 (2), 167 (3), 73 (61), 61 (100);

HRMS found 516.1011 ($M^{\cdot+}$), $C_{22}H_{29}O_6I$ requires 516.1009.

Anal. Found: C, 51.37; H, 6.05. Calcd for $C_{22}H_{29}O_6I$: C, 51.17; H, 5.66.

Preparation of the target compound <u>135</u> based on selenoxide elimination



$NaBH_4$ (4 mg, 0.106 mmol) was added to a suspension of *o*-nitrophenyl selenocyanate[80] (20 mg, 0.088 mmol) in dry EtOH (0.8 ml) with vigorous stirring. An evolution of HCN occurred and the colour changed from light yellow to dark red indicating the presence of *o*-nitrophenylselenide anions. This solution was added dropwise to a solution of iodide **<u>130</u>** (15 mg, 0.029 mmol) in EtOH (1.5 ml) and the reaction mixture was stirred at room temperature. After 6 hours, a yellow precipitate had formed, which was attributed to the oxidative formation of the diselenide[80]. Nitrogen atmosphere was therefore replaced with argon and $NaBH_4$ (3 mg,

0.079 mmol) was added. When TLC analysis indicated that the reaction was complete (*ca* 48 hours), the solution was diluted with EtOAc, subjected to standard work-up and the solvent removed. Chromatography on silica gel (EtOAc/hexane 1:1) afforded in order of elution:

16-*epi-ent*-10β-Hydroxy-12α-methoxymethoxy-17-(*o*-aminophenylseleno)-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (**133**, 5 mg, 31%):

^{1}H NMR (300 MHz, $CDCl_3$) 7.52 (1H, br d, J = 7.6 Hz, -$C_6\underline{H}_4NH_2$), 7.12 (1H, br t, J = 7.6 Hz, -$C_6\underline{H}_4NH_2$), 6.78 (1H, br d, J = 7.6 Hz, -$C_6\underline{H}_4NH_2$), 6.63 (1H, br t, J = 7.6 Hz, -$C_6\underline{H}_4NH_2$), 4.61 (1H, d, J = 6.9 Hz, -O$\underline{CH_2}OCH_3$), 4.57 (1H, d, J = 6.9 Hz, -O$\underline{CH_2}OCH_3$), 3.71 (1H, m overlapped, H12α), 3.69 (3H, s overlapped, -$CO_2\underline{CH_3}$), 3.30 (3H, s, -$OCH_2O\underline{CH_3}$), 2.83 (1H, d, $J_{6,5}$ = 8.2 Hz, H6), 2.58 (2H, m overlapped, H17), 2.50 (1H, dd overlapped, $J_{11\alpha,11\beta}$ = 14.4 Hz, $J_{11\alpha,12\alpha}$ = 10.1 Hz, H11α), 2.16 (1H, m, H13), 1.41 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.6 Hz, H14α), 1.10 (3H, s, H18);

LRMS 561 ($M^{\cdot+}$, 15), 389 (39), 357 (16), 327 (100), 295 (32), 267 (49), 223 (24), 195 (17), 173 (25), 149 (36);

HRMS found 561.1627 ($M^{\cdot+}$), $C_{28}H_{35}O_6N^{80}Se$ requires 561.1630;

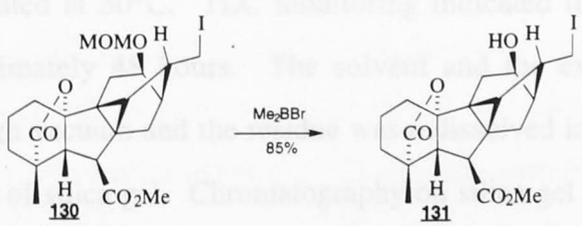
16-*epi-ent*-10β-Hydroxy-12α-methoxymethoxy-17-(*o*-nitrophenylseleno)-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (**132**, 8 mg, 47%):

^{1}H NMR (300 MHz, $CDCl_3$) 8.29 (1H, d, J = 8.1 Hz, -$C_6\underline{H}_4NO_2$), 7.52 (2H, m, -$C_6\underline{H}_4NO_2$), 7.32 (1H, m, -$C_6\underline{H}_4NO_2$), 4.60 (1H, d, J = 6.8 Hz, -O$\underline{CH_2}OCH_3$), 4.55 (1H, d, J = 6.8 Hz, -O$\underline{CH_2}OCH_3$), 3.73 (1H, m overlapped, H12α), 3.70 (3H, s overlapped, -$CO_2\underline{CH_3}$), 3.31 (3H, s, -$OCH_2O\underline{CH_3}$), 2.89 (1H, d, $J_{6,5}$ = 8.7 Hz, H6), 2.78 (2H, d, J = 8.1 Hz, H17), 2.51 (1H, dd overlapped, H11α), 2.50 (1H, d, $J_{5,6}$ = 8.7 Hz, H5), 2.20 (1H, m, H13), 1.50 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 12.4 Hz, H14α), 1.11 (3H, s, H18).

The *o*-nitroseleno derivative **132** (8 mg, 0.014 mmol) was dissolved in dry CH_2Cl_2 (1 ml) under an argon atmosphere. The solution was cooled to -78°C in an acetone/dry ice bath and treated with Me_2BBr (1 drop added *via* a glass pipette). After the solution had been stirred at -78°C for 5 minutes, a 1M aqueous $NaHCO_3$ (0.5 ml) was added at this temperature and the resulting slurry was allowed to warm up to room temperature with efficient stirring. The mixture was then transferred into a separating funnel and washed with aqueous 1M $NaHCO_3$. The organic layer was separated and the inorganic phase extracted with 3 x 1 ml of CH_2Cl_2. The combined organic extracts were dried, the solvent removed and the residue redissolved in THF (0.8 ml) and 30% H_2O_2 (50 µl). TLC analysis after 14 hours showed that only the highly polar selenoxide (**134**) was present in the reaction mixture. Et_3N (100 µl) was added and the solution was heated at 50°C. When TLC revealed that the elimination was complete (*ca* 4 hours), the reaction mixture was directly chromatographed on silica gel (EtOAc/hexane 1:1) to afford olefin **135** (4 mg, 80% from **132**):

^{1}H NMR (300 MHz, $CDCl_3$) 5.01 (1H, s, H17), 4.90 (1H, s, H'17), 3.87 (1H, m, H12α), 3.72 (3H, s, $-CO_2\underline{CH_3}$), 2.89 (1H, d, $J_{6,5}$ = 8.9 Hz, H6), 2.65 (1H, dd, $J_{11\alpha,11\beta}$ = 14.8 Hz, $J_{11\alpha,12\alpha}$ = 9.2 Hz, H11α), 2.41 (1H, m, H13), 2.12 (1H, d, $J_{5,6}$ = 8.9 Hz, H5), 2.04 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 11.9 Hz, $J_{14\beta,13}$ = 6.2 Hz, H14β), 2.02 (1H, s overlapped, H15), 1.65 (1H, dd overlapped, $J_{11\beta,11\alpha}$ = 14.8 Hz, $J_{11\beta,12\alpha}$ = 3.8 Hz, H11β), 1.63 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 11.9 Hz, H14α), 1.09 (3H, s, H18).

16-*epi*-*ent*-10β,12α-Dihydroxy-17-iodo-20-nor-9α,15α-cyclogibberellane-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (131)

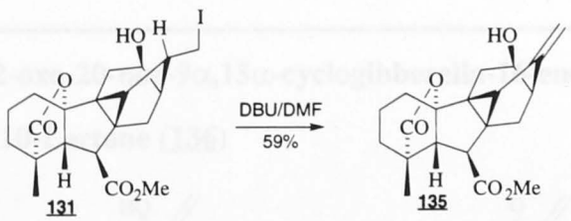


Compound **130** (30 mg, 0.058 mmol) was dissolved in dry CH_2Cl_2 (2 ml) under an argon atmosphere. The solution was cooled to -78°C and treated with Me_2BBr

(approximately 17 µl, 0.174 mmol) which was added dropwise *via* a glass pipette. After the solution was stirred at -78°C for 5 minutes, a 1M aqueous $NaHCO_3$ (0.5 ml) was added at this temperature and the resulting slurry was allowed to warm up to room temperature with efficient stirring. The mixture was then transferred into a separating funnel and washed with 1M aqueous $NaHCO_3$. The organic layer was separated and the inorganic phase extracted with 3 x 2 ml of CH_2Cl_2. Combined organic layers were dried, concentrated under reduced pressure and chromatographed on silica gel (EtOAc/hexane 2:3) to afford the deprotected alcohol **131** (23 mg, 85%) as a solid:

^{1}H NMR (300 MHz, $CDCl_3$) 3.91 (1H, m, H12α), 3.71 (3H, s, -$CO_2\underline{CH_3}$), 3.03 (1H, m, H17), 2.95 (1H, m, H'17), 2.85 (1H, d, $J_{6,5}$ = 9.0 Hz, H6), 2.59 (1H, m overlapped, H13), 2.51 (1H, dd overlapped, $J_{11\alpha,11\beta}$ = 14.4 Hz, $J_{11\alpha,12\alpha}$ = 10.8 Hz, H11α), 2.08 (1H, d overlapped, $J_{5,6}$ = 9.0 Hz, H5), 1.92 (1H, dd, $J_{14\beta,14\alpha}$ = 12.6 Hz, $J_{14\beta,13}$ = 5.0 Hz, H14β), 1.70 (1H, dd, $J_{11\beta,11\alpha}$ = 14.4 Hz, $J_{11\beta,12\alpha}$ = 4.8 Hz, H11β), 1.45 (1H, d, $J_{14\alpha,14\beta}$ = 12.6 Hz, H14α), 1.11 (3H, s, H18).

***ent*-10β,12α-Dihydroxy-20-nor-9α,15α-cyclogibberella-16-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (135)**



Iodide **131** was dried under high vacuum for 2 hours and the flask was charged with an argon atmosphere. Dry DMF (2 ml) was added followed by DBU (150 µl) and the solution was heated at 50°C. TLC monitoring indicated that the reaction was complete in approximately 48 hours. The solvent and the excess of DBU were evaporated under high vacuum and the residue was redissolved in EtOAc and filtered through a short plug of silica gel. Chromatography on silica gel (EtOAc/hexane 1:1) afforded olefin **135** (10 mg, 59%) as an oil:

IR ($CDCl_3$) ν_{max} 3600, 3560, 2960, 1770, 1730, 1440, 1380, 1270, 1200, 1130, 1060 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 5.01 (1H, s, H17), 4.90 (1H, s, H'17), 3.87 (1H, m, H12α), 3.72 (3H, s, -$CO_2\underline{CH_3}$), 2.89 (1H, d, $J_{6,5}$ = 8.9 Hz, H6), 2.65 (1H, dd, $J_{11\alpha,11\beta}$ = 14.8 Hz, $J_{11\alpha,12\alpha}$ = 9.2 Hz, H11α), 2.41 (1H, m, H13), 2.12 (1H, d, $J_{5,6}$ = 8.9 Hz, H5), 2.04 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 11.9 Hz, $J_{14\beta,13}$ = 6.2 Hz, H14β), 2.02 (1H, s overlapped, H15), 1.65 (1H, dd overlapped, $J_{11\beta,11\alpha}$ = 14.8 Hz, $J_{11\beta,12\alpha}$ = 3.8 Hz, H11β), 1.63 (1H, d overlapped, $J_{14\alpha,14\beta}$ = 11.9 Hz, H14α), 1.09 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 178.6 (CO), 172.7 (CO), 146.2 (C16), 106.9 (C17), 92.9 (C10), 68.5 (C12), 56.4 (C5), 52.2 (-$CO_2\mathbf{C}H_3$), 48.0 (C4), 47.4 (C6), 45.7 (C13), 41.4 (C8), 37.4 (C9), 35.0 (C14), 30.8 (C11), 30.2 (C15), 28.4 (CH_2), 27.2 (CH_2), 19.1 (CH_2), 16.7 (C18);

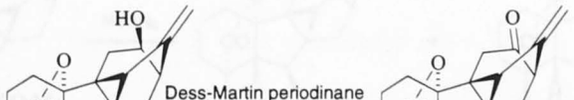
LRMS 344 ($M^{\cdot+}$, 14), 326 ($M^{\cdot+}$-H_2O, 4), 312 (22), 300 (20), 285 (24), 266 (16), 256 (39), 254 (98), 240 (100), 223 (16), 211 (15), 197 (62), 181 (22), 141 (21);

HRMS found 344.1622 ($M^{\cdot+}$), $C_{22}H_{24}O_5$ requires 344.1624.

GC-MS (12-OTMS) 416 ($M^{\cdot+}$, 100), 401 (15), 372 (13), 357 (23), 326 (33), 313 (38), 300 (8), 282 (8), 267 (14), 254 (26), 240 (30), 223 (47), 196 (26), 181 (12);

KRI (12-OTMS) 2487.

***ent*-10β-Hydroxy-12-oxo-20-nor-9α,15α-cyclogibberella-16-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>136</u>)**



Alcohol **<u>135</u>** (10.5 mg, 0.031 mmol) was dissolved in dry dichloromethane (1 ml). Dess-Martin periodinane (29 mg, 0.068 mmol) was added to the solution and the resulting mixture was stirred overnight. The cloudy solution was poured into a separating funnel and washed successively with aqueous $Na_2S_2O_3$ and $KHCO_3$ until the organic layer became clear. The combined inorganic washings were then extracted with CH_2Cl_2 (3 x 5 ml). Combined organic extracts were dried, concentrated under

reduced pressure and chromatographed on silica gel (EtOAc/hexane 3:7) to afford ketone **136** (9 mg, 87%) as a solid:

IR ($CDCl_3$) ν_{max} 2960, 1770, 1730, 1440, 1380, 1280, 1260, 1200, 1170, 1130, 980 cm^{-1};

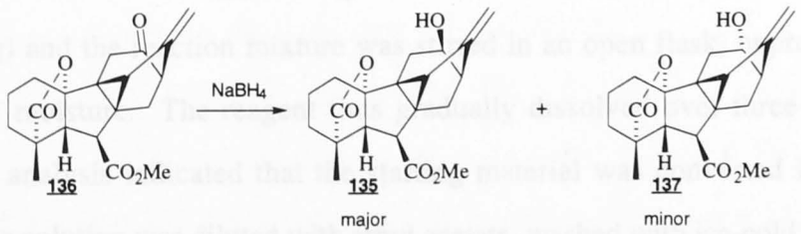
^{1}H NMR (300 MHz, $CDCl_3$) 5.04 (1H, s, H17), 5.02 (1H, s, H'17), 3.75 (3H, s, $-CO_2\underline{CH_3}$), 3.06 (1H, d, $J_{13,14\beta}$ = 5.8 Hz, H13), 2.97 (1H, d, $J_{6,5}$ = 8.9 Hz, H6), 2.85 (1H, d, $J_{11\alpha,11\beta}$ = 19.8 Hz, H11α), 2.64 (1H, d, $J_{11\beta,11\alpha}$ = 19.8 Hz, H11β), 2.27 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 12.5 Hz, $J_{14\beta,13}$ = 5.8 Hz, H14β), 2.24 (1H, d overlapped, $J_{5,6}$ = 8.9 Hz, H5), 2.21 (1H, s, H15), 1.99 (1H, d, $J_{14\alpha,14\beta}$ = 12.5 Hz, H14α), 1.12 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 204.9 (C12), 178.1 (CO), 172.3 (CO), 143.4 (C16), 109.3 (C17), 92.1 (C10), 56.8 (C5), 56.7 (C6), 52.4 ($-CO_2\mathbf{C}H_3$), 48.1 (C4), 45.4 (C13), 40.8 (C8), 35.0 (C9), 34.4 (C14), 32.4 (C11), 30.1 (CH_2), 28.9 (C15), 27.5 (CH_2), 19.1 (CH_2), 16.6 (C18);

LRMS 342 ($M^{\cdot+}$, 100), 314 (7), 299 (36), 283 (33), 268 (16), 255 (77), 240 (99), 227 (17), 211 (68), 196 (67), 181 (38), 167 (43), 155 (48), 141 (38), 129 (27);

HRMS found 342.1469 ($M^{\cdot+}$), $C_{22}H_{22}O_5$ requires 342.1467.

Reduction of ketone 136



$NaBH_4$ (0.13 mg in 30 µl of MeOH, 3.5 µmol) was added to the solution of ketone **136** (0.8 mg, 2.34 µmol) and the reaction mixture was stirred for 5 minutes at room temperature. 2 drops of aqueous 1M HCl were added to destroy the excess of the reagent and the solution was diluted with EtOAc, subjected to standard work-up, dried and the solvent evaporated under reduced pressure. GC-MS analysis of the residue identified in order of elution:

***ent*-10β,12β-Dihydroxy-20-nor-9α,15α-cyclogibberella-16-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**137**, minor component):

GC-MS (12-OTMS) 416 ($M^{\cdot+}$, 100), 401 (7), 385 (4), 369 (13), 357 (16), 326 (29), 313 (39), 298 (7), 281 (11), 267 (12), 254 (26), 240 (28), 223 (43), 196 (24), 181 (13);

KRI (12-OTMS) 2458;

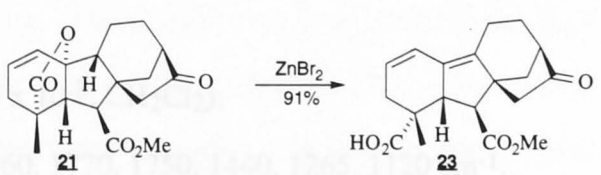
***ent*-10β,12α-Dihydroxy-20-nor-9α,15α-cyclogibberella-16-ene-7,19-dioic Acid 7-(Methyl Ester) 19,10-Lactone** (**135**, major component):

GC-MS (12-OTMS) 416 ($M^{\cdot+}$, 100), 401 (15), 372 (13), 357 (23), 326 (33), 313 (38), 300 (8), 282 (8), 267 (14), 254 (26), 240 (30), 223 (47), 196 (26), 181 (12);

KRI (12-OTMS) 2487.

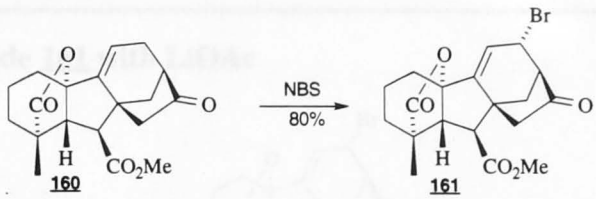
5.4 CHAPTER 4 EXPERIMENTAL

***ent*-16-Oxo-17,20-dinorgibberella-1,9-diene-7,19-dioic Acid 7-(Methyl ester)** (**23**)



A solution of ketone **21** (300mg, 0.909mmol) in Et_2O (40ml) was treated with $ZnBr_2$ (3.5g) and the reaction mixture was stirred in an open flask, unprotected from atmospheric moisture. The reagent thus gradually dissolved over three hours, after which TLC analysis indicated that the starting material was converted into a single product. The solution was diluted with ethyl acetate, washed with ice-cold aqueous 1M HCl (1x), brine (2x), dried and the solvent removed under reduced pressure. Chromatography on silica gel (EtOAc/hexane 1:1, plus 1% vol. AcOH) furnished diene acid **23** (273mg, 91%), identical with the previously prepared material[36].

***ent*-12β-Bromo-10β-hydroxy-16-oxo-17,20-dinorgibberella-9(11)-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (161)**



Enone **160** (73 mg, 0.221mmol) was dissolved in dry CCl_4 (5 ml) under argon, N-bromosuccinimide (65 mg, 0.376 mmol, freshly crystallised), added and the reaction mixture brought to reflux. A solution of dibenzoylperoxide (approximately 0.5 mg in 50 µl of CCl_4) was added *via* syringe. When the mixture turned pale yellow (approximately 35 min.), TLC monitoring revealed that the reaction was complete. The mixture was cooled, diluted with EtOAc, washed with saturated $Na_2S_2O_3$, $KHCO_3$ and brine. The organic phase was dried and concentrated under reduced pressure. Chromatography on silica gel (hexane/EtOAc 65:35) afforded pure bromide **161** (72 mg, 80%) as a solid, which crystallised from Et_2O:

mp 123-125°C;

$[\alpha]_D^{20}$ + 191.4° (c 25 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2960, 1770, 1750, 1440, 1265, 1120 cm^{-1}.

^{1}H NMR (300 MHz, $CDCl_3$) 6.01 (1H, dd, $J_{11,12\beta}$ = 3.8 Hz, $J_{11,13}$ = 1.1 Hz, H11), 4.73 (1H, dd, $J_{12\beta,11}$ = 3.8 Hz, $J_{12\beta,13}$ = 2.7 Hz, H12β), 3.76 (3H, s, -$CO_2\underline{CH_3}$), 3.10 (1H, m, H13), 2.82 (1H, d, $J_{5,6}$ = 11.4 Hz, H5), 2.52 (1H, d, $J_{6,5}$ = 11.4 Hz, H6), 2.41 (2H, m), 2.22 (1H, dd overlapped, J_1 = 14.4 Hz, J_2 = 4.8 Hz), 2.17 (1H, d overlapped, $J_{15,15'}$ = 17.3 Hz, H15), 1.94 (1H, d overlapped, $J_{15',15}$ = 17.3 Hz, H'15), 1.13 (3H, s, H18);

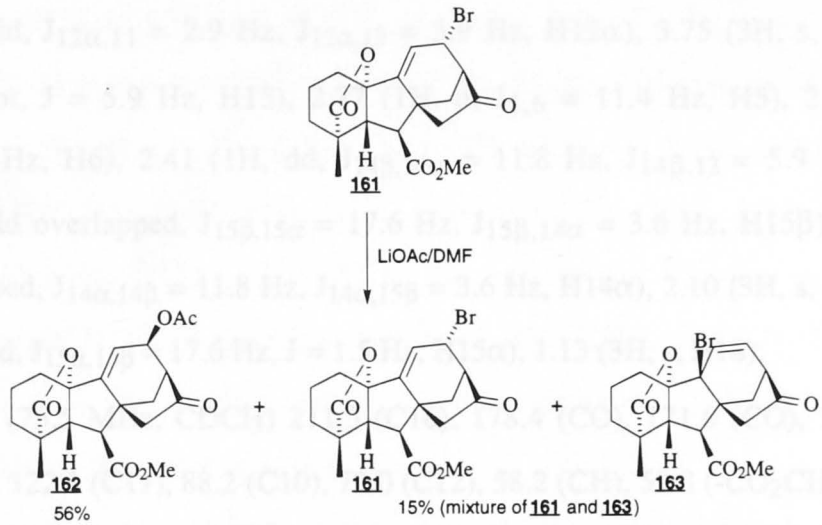
^{13}C NMR (75.5 MHz, $CDCl_3$) 212.0 (C16), 178.4 (CO), 171.6 (CO), 149.1 (C9), 125.0 (C11), 88.2 (C10), 57.6 (CH), 55.5 (CH), 52.8 (-$CO_2\mathbf{C}H_3$), 51.6 (e), 49.9 (CH), 48.7 (e), 48.6 (e), 45.7 (CH), 35.1 (CH_2), 34.9 (CH_2), 30.3 (CH_2), 19.7 (CH_2), 17.4 (C18);

LRMS 329 ($M^{\cdot+}$ - Br, 15), 287 (83), 241 (23), 227 (43), 199 (34), 183 (98), 141 (68), 80 (100), 55 (49);

HRMS found 329.1388 ($M^{\cdot+}$ - Br), $C_{19}H_{21}O_5$ requires 329.1389.

Anal. Found: C, 56.07; H, 4.95. Calcd for $C_{19}H_{21}O_5Br$: C, 55.76; H, 5.17.

Reaction of bromide 161 with LiOAc



Bromocompound **161** (65 mg, 0.159 mmol) was dissolved in dry DMF under argon. Dry LiOAc (500 mg, 7.58mmol) was added and the reaction mixture was stirred at 45°C for 60 hours. The solvent was evaporated under high vacuum, the residue suspended in EtOAc and washed with brine (2x). The organic phase was dried and concentrated under reduced pressure. Chromatography on silica gel (EtOAc/hexane 2:3) gave in order of elution:

a mixture of the starting material and ***ent*-9α-Bromo-10β-hydroxy-16-oxo-17,20-dinorgibberella-11-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** (**163**, 10 mg, 15%):

^{1}H NMR (300 MHz, $CDCl_3$) 6.27 (1H, d, J = 8.5 Hz, H11), 6.02 (1H, dd, $J_{12,11}$ = 8.5 Hz, $J_{12,13}$ = 6.9 Hz, H12), 3.75 (3H, s, $-CO_2\underline{CH_3}$), 2.94 (1H, d, $J_{5,6}$ = 9.0 Hz, H5), 1.41 (1H, d, $J_{14\alpha,14\beta}$ = 11.7 Hz, H14α), 1.14 (3H, s, H18) (**163**);

***ent*-12α-Acetoxy-10β-hydroxy-16-oxo-17,20-dinorgibberella-9(11)-ene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone** as an oil (**162**, 35 mg, 56%), which crystallised from Et_2O:

mp 199-201°C

$[\alpha]_D^{20}$ + 54° (c 33 x 10^{-3}, CH_2Cl_2);

IR ($CDCl_3$) ν_{max} 2860, 1720, 1700, 168, 1325, 1185, 1000 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 5.78 (1H, dd, $J_{11,12\alpha}$ = 2.9 Hz, $J_{11,13}$ = 1.2 Hz, H11), 5.64 (1H, dd, $J_{12\alpha,11}$ = 2.9 Hz, $J_{12\alpha,13}$ = 5.9 Hz, H12α), 3.75 (3H, s, -CO_2CH_3), 3.04 (1H, bt, J = 5.9 Hz, H13), 2.77 (1H, d, $J_{5,6}$ = 11.4 Hz, H5), 2.61 (1H, d, $J_{6,5}$ = 11.4 Hz, H6), 2.41 (1H, dd, $J_{14\beta,14\alpha}$ = 11.8 Hz, $J_{14\beta,13}$ = 5.9 Hz, H14β), 2.29 (1H, dd overlapped, $J_{15\beta,15\alpha}$ = 17.6 Hz, $J_{15\beta,14\alpha}$ = 3.6 Hz, H15β), 2.18 (1H, dd overlapped, $J_{14\alpha,14\beta}$ = 11.8 Hz, $J_{14\alpha,15\beta}$ = 3.6 Hz, H14α), 2.10 (3H, s, -OCO<u>CH_3</u>), 1.99 (1H, dd, $J_{15\alpha,15\beta}$ = 17.6 Hz, J = 1.5 Hz, H15α), 1.13 (3H, s, H18);

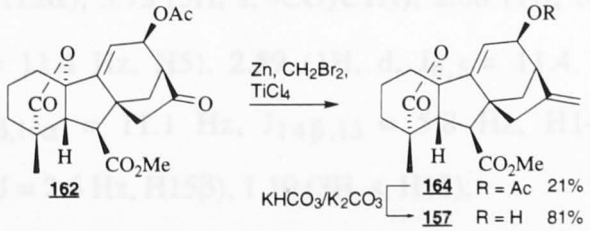
^{13}C NMR (75.5 MHz, $CDCl_3$) 211.5 (C16), 178.4 (CO), 171.6 (CO), 170.6 (CO), 152.2 (C9), 122.1 (C11), 88.2 (C10), 72.0 (C12), 58.2 (CH), 52.8 (-CO_2**C**H_3), 51.3 (e), 50.1 (CH), 50.0 (e), 48.7 (e), 39.6 (CH_2), 35.1 (CH_2), 30.2 (CH_2), 21.4 (-OCO**C**H_3), 19.7 (CH_2), 17.3 (C18);

LRMS 388 (M^+, 4), 346 (9), 303 (24), 243 (39), 199 (54), 183 (20), 141 (25), 91 (28), 55 (100);

HRMS found 388.1522 (M^+), $C_2H_{24}O_7$ requires 388.1522.

Anal. Found: C, 64.62; H, 6.37. Calcd for $C_{21}H_{24}O_7$: C, 64.94; H, 6.23.

***ent*-12α,10β-Dihydroxy-20-norgibberella-9(11),16-diene-7,19-dioic Acid 7-(Methyl ester) 19,10-Lactone (<u>157</u>)**



Ketone **<u>162</u>** (24 mg, 0.062 mmol) was dissolved in dry THF (1.5 ml) and the solution was treated with freshly prepared Lombardo-Oshima reagent (see page 13) under nitrogen. The reagent was added dropwise in portions until TLC indicated that only a small amount of the starting material was left. The reaction was then quenched

with an aqueous solution of $NaHCO_3$ and the resulting mixture was stirred for 5 minutes. The reaction mixture was diluted with Et_2O, washed with brine, dried and concentrated under reduced pressure. Chromatography on silica gel (EtOAc/hexane 1:3) gave acetate **164** (5mg, 21%) as an oil:

^{1}H NMR (300 MHz, $CDCl_3$) 5.67 (1H, dd, $J_{11,12\alpha}$ = 2.7 Hz, $J_{11,13}$ = 1.5 Hz, H11), 5.61 (1H, dd, $J_{12\alpha,13}$ = 4.9 Hz, $J_{12\alpha,11}$ = 2.7 Hz, H12α), 5.05 (1H, bs, H17), 5.0 (1H, bs, H'17), 3.73 (3H, s, - $CO_2\underline{CH_3}$), 3.11 (1H, bt, J = 4.9 Hz, H13), 2.75 (1H, $J_{5,6}$ = 11.4 Hz, H5), 2.60 (1H, d, $J_{6,5}$ = 11.4 Hz, H6), 2.37 (1H, br d, $J_{15\beta,15\alpha}$ = 15.3 Hz, H15β), 2.19 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 11.2 Hz, $J_{14\beta,13}$ = 6.0 Hz, H14β), 2.10 (3H, s overlapped, -OCO$\underline{CH_3}$), 2.08 (1H, d overlapped, $J_{15\alpha,15\beta}$ = 15.3 Hz, H15α), 1.94 (1H, dd overlapped, $J_{14\alpha,14\beta}$ = 11.2 Hz, $J_{14\alpha,15\beta}$ = 2.6 Hz, H14α), 1.10 (3H, s, H18).

The acetate (5 mg, 0.013 mmol) was dissolved in MeOH (0.8 ml). An aqueous $K_2CO_3/KHCO_3$ solution (20 µl; 125 g of K_2CO_3 and 2 g of $KHCO_3$ in 25 ml of H_2O) was added and the reaction mixture was stirred at room temperature for 5 hours. The solution was then diluted with EtOAc, washed with brine, the organic phase dried and concentrated under reduced pressure. Chromatography on silica gel (EtOAc/hexane 3:7) afforded alcohol **157** (3.6 mg, 81% from **164**) as an oil:

IR ($CDCl_3$) ν_{max} 3540, 2950, 1770, 1730, 1440, 1265, 1130 cm^{-1};

^{1}H NMR (300 MHz, $CDCl_3$) 5.70 (1H, dd, $J_{11,12\alpha}$ = 2.7 Hz, $J_{11,13}$ = 1.3 Hz, H11), 5.16 (1H, bs, H17), 5.10 (1H, bs, H'17), 4.50 (1H, ddd, J = 11.9 Hz, $J_{12\alpha,13}$ = 5.7 Hz, $J_{12\alpha,11}$ = 2.7 Hz, H12α), 3.73 (3H, s, -CO_2CH_3), 2.88 (1H, bt, J = 5.1 Hz, H13), 2.74 (1H, d, $J_{5,6}$ = 11.4 Hz, H5), 2.59 (1H, d, $J_{6,5}$ = 11.4 Hz, H6), 2.19 (1H, dd overlapped, $J_{14\beta,14\alpha}$ = 11.1 Hz, $J_{14\beta,13}$ = 5.8 Hz, H14β), 2.09 (1H, dt, $J_{15\beta,15\alpha}$ = 15.9 Hz, J = 2.6 Hz, H15β), 1.10 (3H, s, H18);

^{13}C NMR (75.5 MHz, $CDCl_3$) 178.5 (CO), 172.0 (CO), 148.6 (C9 or C16), 147.3 (C9 or C16), 125.7 (C11), 111.7 (C17), 88.5 (C10), 70.7 (C12), 58.1 (C5), 53.0 (C8), 52.2 ($CO_2\mathbf{C}H_3$), 49.0 (C6 or C13), 48.4 (C4), 48.1 (C6 or C13), 43.5 (C15), 41.6 (C14), 34.9 (C3), 30.1 (C1), 19.5 (C2), 17.0 (C18);

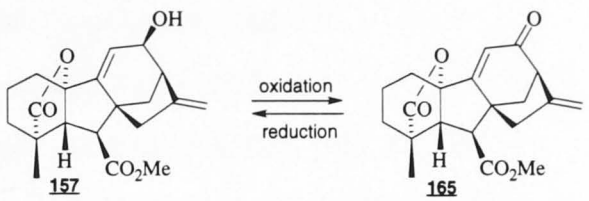
LRMS 344 (M^+, 7), 326 (5), 300 (14), 209 (16), 188 (34), 149 (27), 88 (28), 61 (100);

HRMS found 344.1622 (M^+), $C_{20}H_{24}O_5$ requires 344:1624.

GC-MS (12-OTMS) 416 (M^+, 14), 401 (9), 385 (12), 372 (100), 357 (10), 329 (8), 313 (14), 300 (7), 282 (12), 267 (17), 253 (7), 241 (9), 223 (18), 129 (17);

KRI 2556.

Oxidation/reduction of alcohol 157



Compound **157** (2 mg, 0.006 mmol) was dissolved in CH_2Cl_2 (0.3 ml) and the solution was treated with Dess-Martin periodinane (5 mg, 0.012 mmol). After being stirred overnight, the reaction mixture was worked up in the usual way (see, for example, **58** → **59**) to afford ketone **165**.

The crude material was dissolved in 0.4M solution of $CeCl_3$ in MeOH (0.3 ml) and treated with $NaBH_4$ (approximately 1 mg, 0.026 mmol) at 0°C. After 5 minutes, the excess of the reagent was destroyed with 1 drop of aqueous 1M HCl, the reaction mixture was diluted with EtOAc, subjected to standard work-up, dried and the solvent evaporated *in vacuo*. ^1H NMR analysis revealed the presence of alcohol **164**, with no trace of the other epimer being detectable.

This material was reoxidized under the above conditions and resubjected to the $NaBH_4$ reduction in MeOH with the exclusion of $CeCl_3$, the outcome being as above.

REFERENCES

1. H. Schraudolf, *Proceedings of the Royal Society of Edinburgh* 1985, **86 B**, 75, and references cited therein.

2. K. Norstog, R. W. Long, *Plant Biology*, Saunders, Philadelphia, 1976, pp. 356-357.

3. L. J. Audus, *Plant Growth Substances*, 3rd edition, Leonard Hill, London, 1973, pp. 301.

4. W. Döpp, *Ber. Deut. Bot. Ges.* 1950, **63**, 139.

5. W. Döpp, *Ber. Deut. Bot. Ges.* 1959, **72**, 11.

6. U. Näf, K. Nakanishi, M. Endo, *Bot. Rev.* 1975, **41**, 315.

7. P. B. Sharp, G. W. Keitt, H. H. Clum, U. Näf, *Physiol. Plant.* 1975, **34**, 101.

8. a) H. Schraudolf, *Nature* 1964, **201**, 98.

   b) B. Voeller, *Science* 1964, **143**, 373.

9. K. Nakanishi, M. Endo, U. Näf, L. F. Johnson, *J. Am. Chem. Soc.* 1971, **93**, 5579.

10. E. J. Corey, A. G. Myers, H. Schraudolf, N. Takahashi, H. Yamane, *Tetrahedron Lett.* 1986, **27**, 5083.

11. E. J. Corey, A. G. Myers, *J. Am. Chem. Soc.* 1985, **107**, 5574.

12. P. R. Zanno, M. Endo, K. Nakanishi, U. Näf, C. Stein, *Naturwissenschaften* 1972, **512**, 12.

13. H. Yamane, K. Nohara, N. Takahashi, H. Schraudolf, *Plant Cell Physiol.* 1987, **28**, 1203.

14. J. E. Nester, S. Veysey, R. C. Coolbaugh, *Planta* 1987, **170**, 26.

15. M. Furber, L. N. Mander, J. E. Nester, N. Takahashi, H. Yamane *Phytochemistry* 1989, **28**, 63.

16. M. Furber, L. N. Mander, *J. Am. Chem. Soc.* 1988, **110**, 4084.

17. H. Yamane, N. Takahashi, K. Takeno, M. Furuya, *Planta* 1979, **147**, 251.

18. H. Yamane, Y. Satoh, K. Nohara, M. Nakayama, N. Murofushi, N. Takahashi, K. Takeno, M. Furuya, M. Furber, L. N. Mander, *Tetrahedron Lett.* 1988, **29**, 3959.

19. P. J. Davies, Ed., *Plant Hormones and Their Role in Plant Growth and Development*, Martinus Nijhoff, Dordrecht, 1987.

20. L. N. Mander, *Chem. Rev.* 1992, **92**, 573.

21. T. Yamauchi, N. Oyama, H. Yamane, N. Ohyama, N. Murofushi, N. Takahashi, H. Schraudolf, M. Furber, L. N. Mander, G. L. Patrick, B. Twitchin, *Phytochemistry* 1991, **30**, 3247.

22. L. N. Mander, personal communication.

23. H. Yamane, personal communication.

24. P. Kraft-Klaunzer, unpublished work.

25. L. N. Mander, H. Yamane, personal communication.

26. P. Gaskin, J. MacMillan, *GC-MS of Gibberellins and Related Compounds: Methodology and a Library of Reference Spectra*, Cantocks Enterprises, Bristol, 1991.

27. M. Furber, L. N. Mander, unpublished work.

28. M. Furber, L. N. Mander, *J. Am. Chem. Soc.* 1987, **109**, 6389.

29. M. Furber, L. N. Mander, G. L. Patrick, *J. Org. Chem.* 1990, **55**, 4860.

30. J. R. Hanson, *Natl. Prod. Rep.* 1990, **7**, 41.

31. L. N. Mander, In *Studies in Natural Product Chemistry*, Atta-ur-Rahman, Ed.; Elsevier, Amsterdam, 1990; pp. 171-212.

32. B. E. Cross, J. F. Grove, A. J. Morrison, *J. Chem. Soc.* 1961, 2498.

33. P. S. Kirkwood, J. MacMillan, M. Sinnott, *J. Chem. Soc., Perkin Trans. 1*, 1980, 2117.

34. E. P. Serebryakov, *Izv. Akad. Nauk SSSR, Ser. Khim.* 1980, 2596.

35. B. M. Fraga, I. Collado, J. R. Hanson, unpublished work.

36. P. Kraft-Klaunzer, M. Furber, L. N. Mander, B. Twitchin, *Tetrahedron Lett.* 1990, **31**, 6235.

37. P. Kraft-Klaunzer, unpublished results.

38. L. N. Mander, personal communication.

39. P. Kraft-Klaunzer, unpublished results.

40. E. J. Corey, R. L. Danheiser, S. Chandrasekaran, G. E. Keck, G. Gopalan, S. D. Larsen, P. Siret, J.-L. Gras, *J. Am. Chem. Soc.* 1978, **100**, 8034.

41. For a review, see H. N. C. Wong, M.-Y. Hon, C.-W. Tse, Y.-C. Yip, J. Tanko, T. Hudlicky, *Chem. Rev.* 1989, **89**, 165.

42. P. Kocovsky, J. Srogl, M. Pour, A. Gogoi, *J. Am. Chem. Soc.* 1994, **116**, 186 and references cited therein.

43. W.-L. A. Chu, L. N. Mander, *Tetrahedron Lett.* 1988, **29**, 2727.

44. W.-L. A. Chu, Ph.D. Thesis, Australian National University, 1988.

45. V. K. Bhaskar, W.-L. A. Chu, P. A. Gaskin, L. N. Mander, N. Murofushi, D. W. Pearce, R. P. Pharis, N. Takahashi, I. Yamaguchi, *Tetrahedron Lett.* 1991, **32**, 6203.

46. L. N. Mander, G. L. Patrick, *Tetrahedron Lett.* 1990, **31**, 423.

47. P. Kraft-Klaunzer, unpublished work.

48. R. D. Dawe, L. N. Mander, J. V. Turner *Tetrahedron Lett.* 1985, **26**, 363.

49. For a discussion of reductive cleavages of vinylcyclopropanes, see S. W. Staley, A. S. Heyn, *Tetrahedron* 1974, **30**, 3671.

50. For reductive fissions of cyclopropyl ketones related to gibberellins, see J. V. Turner, B. F. Anderson, L. N. Mander, *Aust. J. Chem.* 1980, **33**, 1061 and references cited therein.

51. P. Kraft-Klaunzer, unpublished results.

52. V. J. Bhaskar, unpublished results.

53. T. W. Greene, P. G. M. Wuts, *Protective Groups in Organic Synthesis*, 2nd edition, John Wiley & Sons, Inc., New York, 1991:

a) pp. 71.

b) pp. 17-20 and references cited therein.

c) pp. 77-83 and references cited therein.

d) pp. 30-31 and references cited therein.

54. L. Lombardo, L. N. Mander, J. V. Turner, *Aust. J. Chem.* 1981, **34**, 745.

55. E. V. Dehmlow, S. S. Dehmlow, *Phase Transfer Catalysis*, VCH publishers, Inc., New York, 1993, pp. 39.

56. F. R. van Heerden, J. J. van Zyl, G. J. H. Rall, E. V. Brandt, D. G. Roux, *Tetrahedron Lett.* 1978, 661.

57. B. Twitchin, personal communication.

58. P. Kraft-Klaunzer, unpublished results.

59. R. D. Dawe, L. N. Mander, J. V. Turner, X.-F. Pan, *Tetrahedron Lett.* 1985, **26**, 5725.

60. T. L. Macdonald, K. J. Natalie, Jr., G. Prasad, J. S. Sawyer, *J. Org. Chem.* 1986, **51**, 1124.

61. M. Furber, L. N. Mander, G. L. Patrick, A. C. Willis, *Acta Cryst.* 1992, **C 48**, 1348.

62. Y. Fujimoto, T. Tatsuno, *Tetrahedron Lett.* 1976, 3325.

63. L. Lombardo, *Tetrahedron Lett.* 1982, **23**, 4293, and references cited therein.

64. R. E. Ireland, L. Liu, *J. Org. Chem.* 1993, **58**, 2899, and references cited therein.

65. S. C. Dolan, J. MacMillan, *J. Chem. Soc. Chem. Commun.* 1985, 1588.

66. D. H. R. Barton, J. Cs. Jaszberenyi, *Tetrahedron Lett.* 1989, **30**, 2619.

67. M. Sherburn, personal communication.

68. For an excellent overview of the synthetic use of borane reagents, see W. Carruthers, *Some Modern Methods of Organic Synthesis*, 3rd edition, Cambridge University Press, 1986, pp. 288-302.

69. V. K. Bhaskar, unpublished results.

70. H. Seto, L. N. Mander, *Syn. Commun.* 1992, **22**, 2823.

71. F. N. Tebbe, G. W. Parshall, G. S. Reddy, J. Am. Chem. Soc. 1978, **100**, 3611.

72. J. M. Tour, P. V. Bedworth, R. Wu, *Tetrahedron Lett.*, 1989, **30**, 3927.

73. J. Ager, *Synthesis* 1984, 384.

74. M. H. Beale, R. Hooley, J. MacMillan, In *Plant Growth Substances 1985*, M. Bopp, Ed.; Springer-Verlag Berlin, 1986; pp. 65.

75. L. F. Fieser, M. Fieser, *Reagents for Organic Synthesis*, Vol. 1, John Wiley & Sons, Inc., New York, 1967; pp. 606-609.

76. For an excellent text describing a number of aspects of modern NMR techniques, see A. E. Derome, *Modern NMR Techniques for Chemistry Research*, Pergamon Press, London, 1987.

77. L. Lerner, A. Bax, *J. Magn. Reson.* 1986, **69**, 375.

78. G. E. Martin, R. C. Crouch, *J. Nat. Prod.* 1991, **54**, 1.

79. J. L. Marshall, *Carbon-Carbon and Carbon-Proton NMR Couplings: Application in Organic Stereochemistry and Conformational Analysis*, A. P. Marchand, Ed.;

Methods in Stereochemical Analysis, Vol. 2; Verlag Chemie International, Deerfield Beach, Florida, 1983.

80. K. B. Sharpless, M. W. Young, *J. Org. Chem.* 1975, **40**, 947.

81. P. A. Grieco, S. Gilman, M. Nishizawa, *J. Org. Chem.* 1976, **41**, 1485.

82. a) For a review, see D. J. Clive, *Tetrahedron* 1978, **34**, 1049.

b) For a list of recent examples of the application of this procedure, see R. C. Larock, *Comprehensive Organic Transformations: A Guide to Functional Group Preparations*, VCH Publishers, Inc., New York, 1989; pp. 149.

83. P. Kocovsky, *Collect. Czech. Chem. Commun.* 1994, **59**, 1, (review) and references cited therein:

a) Discussion of factors governing electrophilic additions: pp. 3-38.

b) Discussion of the reactivity of various groups as internal nucleophiles: pp. 21-23.

84. C. L. Willis, *Tetrahedron Lett.* 1987, **28**, 6705.

85. P. J. Garegg, B. Samuelsson, *J. Chem. Soc., Perkin Trans. 1*, 1980, 2866.

86. Y. Guindon, C. Yoakim, H. E. Morton, *J. Org. Chem.* 1984, **49**, 3912.

87. For the use of an amine base to promote the decomposition of selenoxides, see H. J. Reich, J. M. Renga, I. L. Reich *J. Am. Chem. Soc.* 1975, **97**, 5434.

88. a) T. Yokota, N. Murofushi, T. Takahashi, S. Tamura *Agric. Biol. Chem.* 1971, **35**, 573.

b) H. Fukui, R. Nemori, K. Koshimizu, Y. Yamazaki, *Agric. Biol. Chem.* 1977, **41**, 181.

89. J. MacMillan, R. J. Pryce, *J. Chem. Soc.* 1967, 740.

90. a) T. Yamauchi, Doctoral Thesis: "Physiological and Organic Chemical Studies on the Fern Antheridiogens", The University of Tokyo, 1994.

b) M. Furber, P. Kraft-Klaunzer, L. N. Mander, M. Pour, T. Yamauchi, N. Murofushi, H. Yamane, H. Schraudolf, *Aust. J. Chem.*, accepted for publication.

91. M. Karplus, *J. Am. Chem. Soc.* 1963, **85**, 2870.

92. H. Yamane, L. N. Mander, D. Owen, *Bioscience, Biotechnology and Biochemistry*, in preparation.

93. W. C. Still, M. Kahn, A. Mitra, *J. Org. Chem.* 1978, **43**, 2923.

94. D. D. Perrin, W. L. F. Armarego, D. R. Perrin, *Purification of Laboratory Chemicals*, Pergamon Press, London, 1986, 2nd edition.

95. D. R. Burfield, R. H. Smithers, *J. Org. Chem.* 1978, **43**, 3966.

96. Authentic sample of compound **46** was kindly provided by Bruce Twitchin.

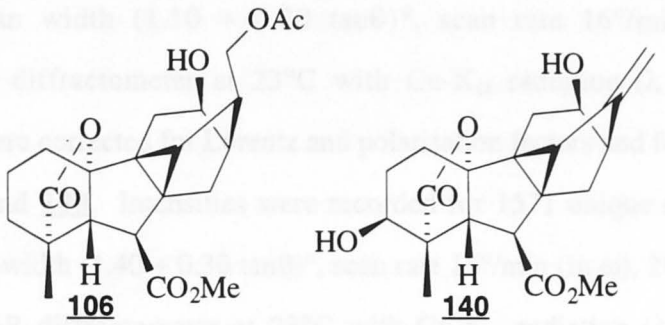
97. Z. J. Duri, J. R. Hanson, *J. Chem. Soc., Perkin Trans. 1*, 1984, 603.

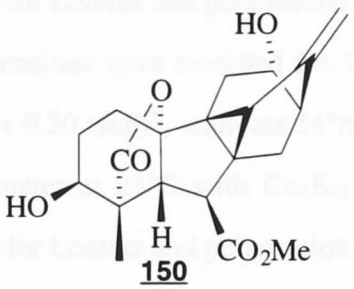
98. The GA_4 derivative **121** was kindly supplied by Bruce Twitchin.

99. S. R. Hall, H. D. Flack, J. M. Stewart, Eds., *XTAL3.2 Reference Manual*, Universities of Western Australia, Geneva and Maryland, Lamb, Perth, 1992.

APPENDIX

Data from Single-Crystal X-Ray Diffraction Analyses of Compounds 106, 140 and 150.





1. Crystal Data

Compound **106**. $C_{22}H_{28}O_7$, M = 404.46, orthorhombic space group $P2_12_12_1$ (#19), a = 10.125(1), b = 10.631(2), c = 19.551(1) Å, V = 2104.4(3) Å^3, Z = 4, d_c = 1.276 g/cm^3. Colourless crystals. Crystal dimensions 0.27 x 0.17 x 0.14 mm, μ[Cu-K$_\alpha$] = 7.80 cm^{-1}.

Compound **140**. $C_{20}H_{24}O_6$, M = 360.41, orthorhombic space group $P2_12_12_1$ (#19), a = 7.279(3), b = 9.415(2), c = 25.724(4) Å, V = 1762.9(8) Å^3, Z = 4, d_c = 1.358 g/cm^3. Colourless prismatic crystals. Crystal dimensions 0.15 x 0.16 x 0.23 mm, μ[Cu-K$_\alpha$] = 7.84 cm^{-1}.

Compound **150**. $C_{20}H_{24}O_6$, M = 360.41, orthorhombic space group $P2_12_12_1$ (#19), a = 10.177(1), b = 12.986(1), c = 13.900(1) Å, V = 1837.0(3) Å^3, Z = 4, d_c = 1.303 g/cm^3. Colourless, block-shaped crystals. Crystal dimensions 0.14 x 0.14 x 0.30 mm, μ[Cu-K$_\alpha$] = 7.53 cm^{-1}.

2. Data Collection and Processing

Compound **106**. Intensities were recorded for 1808 unique reflections by an ω-2θ scan, scan width $(1.10 + 0.30 \tan\theta)°$, scan rate 16°/min (in ω) on a Rigaku-AFC6R diffractometer at 23°C with Cu-K_α radiation (λ = 1.54178 Å). Intensity data were corrected for Lorentz and polarisation factors and for absorption.

Compound **140**. Intensities were recorded for 1571 unique reflections by an ω-2θ scan, scan width $(1.40 + 0.30 \tan\theta)°$, scan rate 16°/min (in ω), $2\theta_{max}$ = 120.2° on a Rigaku-AFC6R diffractometer at 23°C with Cu-K_α radiation (λ = 1.54178 Å). Intensity data were corrected for Lorentz and polarisation factors and for absorption.

Compound **150**. Intensities were recorded for 1609 unique reflections by an ω-2θ scan, scan width $(1.00 + 0.30 \tan\theta)°$, scan rate 16°/min (in ω), $2\theta_{max}$ = 120.1° on a Rigaku-AFC6R diffractometer at 23°C with Cu-K_α radiation (λ = 1.54178 Å). Intensity data were corrected for Lorentz and polarisation factors and for absorption.

3. Structure analysis and refinement

Compound **106**. The structure was solved by direct methods and expanded using Fourier techniques. The non-hydrogen atoms were refined anisotropically. Hydrogen atoms attached to carbon atoms were included in calculated positions and their parameters were not refined; the hydrogen on O(4) was located in a difference map and its positional parameters were refined in the least-squares process. It was apparent at this stage that there was some disorder of the 17-acetoxy group. Refinement of this section of the molecule required the use of restraints in the crystallographic least squares refinement, and XTAL3.2 programs[99] were subsequently used. Hydrogen atom parameters were not refined, but were regularly calculated. The absolute configuration of the molecule was assigned on the basis of the known chirality of centres in its chemical precursors. The final cycle of full-matrix least-squares refinement was based on 1578 observed reflections ($I > 3.00\sigma(I)$), 275 variable parameters and 14 restraints and converged with unweighted and weighted agreement factors of R = 0.051 and R_w = 0.070. The maximum and minimum peaks on the final difference Fourier map corresponded to 0.31(3) and -0.24 $e^-/Å^3$, respectively. All

calculations were performed using the teXsan crystallographic software package of Molecular Structure Corporation. The full set of data including bond lenghts and valence angles for the non-hydrogen atoms, anisotropic thermal parameters and atomic parameters, together with their estimated standard deviations, have been deposited at Cambridge Crystallographic Data Centre.

Compound **140**. The structure was solved by direct methods and expanded using Fourier techniques. The non-hydrogen atoms were refined anisotropically. Hydrogen atoms attached to carbon atoms were included in calculated positions and their parameters were not refined; those on oxygen atoms were located in a difference map and their positional parameters were refined in the least-squares process. The absolute configuration of the molecule was assigned on the basis of the known chirality of centres in its chemical precursors. The final cycle of full-matrix least-squares refinement was based on 1413 observed reflections ($I > 3.00\sigma(I)$) and 241 variable parameters and converged with unweighted and weighted agreement factors of $R = 0.032$ and $R_w = 0.031$. The maximum and minimum peaks on the final difference Fourier map corresponded to 0.13 and -0.14 $e^-/Å^3$, respectively. All calculations were performed using the teXsan crystallographic software package of Molecular Structure Corporation. The full set of data including bond lenghts and valence angles for the non-hydrogen atoms, anisotropic thermal parameters and atomic parameters, together with their estimated standard deviations, have been deposited at Cambridge Crystallographic Data Centre.

Compound **150**. The structure was solved by direct methods and expanded using Fourier techniques. The non-hydrogen atoms were refined anisotropically. Hydrogen atoms attached to carbon atoms were included in calculated positions and their parameters were not refined; those on oxygen atoms were located in a difference map and their positional parameters were refined in the least-squares process. The absolute configuration of the molecule was assigned on the basis of the known chirality of centres in its chemical precursors. The final cycle of full-matrix least-squares refinement was based on 1441 observed reflections ($I > 3.00\sigma(I)$) and 241 variable parameters and converged with unweighted and weighted agreement factors of

$R = 0.036$ and $R_w = 0.031$. The maximum and minimum peaks on the final difference Fourier map corresponded to 0.16 and -0.18 $e^-/Å^3$, respectively. All calculations were performed using the teXsan crystallographic software package of Molecular Structure Corporation. The full set of data including bond lenghts and valence angles for the non-hydrogen atoms, anisotropic thermal parameters and atomic parameters, together with their estimated standard deviations, have been deposited at Cambridge Crystallographic Data Centre.